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CATEGORIES OF SPECIES NAMES IN ZOOLOGY

By Dr. HOBART M. SMITH

UNIVERSITY OF ROCHESTER

IT is a remarkable fact that zoological taxonomists have persisted for many years in maintaining an oversimplified classification of names for animals, in spite of the diverse kinds of names revealed by the Rules and Opinions of the International Commission on Zoological Nomenclature. For nearly two hundred years taxonomists have commonly referred to species names, or subspecies names, or names not qualifying as either, with rare reference to the fact that these names can differ widely in the application of rules to them. Only one category of the several that exist has been popularly recognized (*nomina nuda*), yet it is clear that not all other names are on a par with each other. Some are valid, some invalid; they may be available or not; there are synonyms and homonyms; some are clearly diagnosed, others not. Names in these and other categories are not treated exactly alike by the rules; each may have a distinct procedure outlined for it. For instance, new names proposed with an acceptable but inadequate diagnosis are to be treated in an almost entirely different manner than names accompanied by adequate diagnoses. In spite of the existence of these several different categories, they have never been clearly defined or named. It is my belief that their clarification will greatly simplify the consideration of taxonomic problems. The present discussion suggests a possible scheme of classification.

I

Briefly, the classification suggested is as follows[1]:

A. *binomialia*[2] (binomial names)
1. *nomina nulla* (taxonomic non-entities; unacceptable names)
   a. *nomina mythologica* (names for mythical species)
   b. *nomina defecta* (unscientific names)
2. *nomina naturalia* (scientific names; taxonomic entities; acceptable names)
   a. *nomina prelinnaeana* (prelinnaean names, unavailable)
   b. *nomina nuda* (nude names; entities without taxonomic standing (unavailable)
   c. *nomina specialia* (species names; entities with taxonomic standing, available)
      1′. *nomina clara* (names at least originally identifiable to species or subspecies by original diagnosis)
         a′. *nomina valida* (valid names)
         b′. *nomina invalida* (invalid names)
            1″. *homonomata*[3] (homonyms)
            2″. *synonomata* (synonyms)
               a″. *synonomata natura* (zoological synonyms)
               b″. *synonomata regula*[4] (nomenclatural synonyms)
      2′. *nomina dubia* (names unidentifiable even originally to species or subspecies by original diagnosis)

II

Binomials are names used in the accepted Linnaean manner, consisting of two categories (one of which may include two terms [species and subspecies]). They need not be acceptable, even though formed in the proper manner. Unacceptable names are *nomina nulla* and belong to the vast reserve of possible *nomina nova*[5]; important among these are *nomina mythologica,* names for mythical species, and *nomina defecta,* names occurring in non-taxonomic literature through error or chance. In taxonomic procedure *nomina nulla* have no significance whatever; they are complete non-entities so far as scientific nomenclature is concerned.

III

Acceptable scientific names are *nomina naturalia;* they are *binomialia* proposed after 1757 in acceptable literature for existing animals or their remains or indications. All such names have special status under the Code. Two groups of names, however, have very little significance; they can not be used as names for species, and in that sense are unavailable. These two groups are *nomina prelinnaeana* and *nomina nuda.* Prelinnaean names have received special comment in Opinion 6, which states in part:

> A pre-Linnaean name, ineligible because of its publication prior to 1758, does not become eligible simply by being cited or reprinted with its original diagnosis after 1757. To become eligible under the Code, such names must be reinforced by adoption or acceptance by the author publishing the reprint. Examples: The citation, subsequent to 1757, of a bibliographic reference to a paper published prior to 1758 does not establish technical names which may appear in said reference; synonymic citation of pre-Linnaean names, as in the tenth edition of Linné's "Systema Naturae," does not establish such names under the Code.

Thus prelinnaean names may be used in postlinnaean literature, without becoming available, unless special indication is given by the author. This is not true of any other names, with the exception noted below.

Nomina nuda are names proposed after 1757 in such a manner as not to fulfill the requirements of Article 25 (see Opinion 126). They heretofore have been considered by the commission as complete non-entities (see Opinions 48, 97, 126, etc.). They would ordinarily be placed among *nomina nulla* in the present classification. In this one respect, of all associations and definitions presented in this article, have I taken the liberty of differing with the commission's implied conclusion. It is apparent that prelinnaean names can be discussed in scientific literature without danger of unwittingly making them available, but this is not so with regard to *nomina nuda,* under the commission's statements to date. They are to be treated exactly like *nomina nova* upon citation subsequent to their first appearance, whether in casual reference, inclusion in synonymy, guesses as to identity or definite adoption as a name. Thus authors are practically denied the right of effectively mentioning *nomina nuda,* which in some cases may have considerable importance. It is my belief that the commission not only should countenance discussion of *nomina nuda* by authors, without automatic availment of the name, but can do so without endangering the norm

[1] I am indebted to Dr. Robert A. McLean, professor of classics at the University of Rochester, for his advice in construction of the terms proposed.

[2] No Greek or Latin word appears to convey the exact meaning which has been associated with the English term binomial (= binary), as opposed to *binominal.* To avoid possible misunderstanding, it is regarded advisable to latinize the English word in the form here cited.

[3] This category may apply also to *nomina dubia.*

[4] This category may apply also to *nomina dubia.*

[5] A newly proposed scientific name (*nomen naturale*) may be termed a *nomen novum* or a *species novum;* it is to be recommended that *species nova* be restricted to new names for newly distinguished species, that the term *nomen novum* in a narrow sense be restricted to new names for "old" species (previously described under other names), and that the latter term in a broad sense be used to cover both categories. It may be recommended also that the terms *nomina nova* and *species nova* be restricted for use only with names qualifying as *nomina naturalia.*

of other procedures. It is here suggested that *nomina nuda* are not to be considered complete non-entities in the field of scientific nomenclature, since they may and do occur in accepted scientific literature subsequent to 1757; the necessity of dealing with them, of discussing them, of determining the status of borderline cases, is apparent. It is an error to regard names that may be so discussed in the same light as mythical names and like names (*nomina nulla*) that have no place in postlinnaean scientific literature; *nomina nuda,* as here defined, are actually a part of that literature, though to be sure the most lowly part. That different action might be incumbent upon this difference in status is not unreasonable; and to deny audience to the special requirements of *nomina nuda* as opposed to *nomina nulla* is to deny the existence of a distinction between the literature of the postlinnaean scientific world and that of either the prelinnaean or the unscientific world.

The recognition of *nomina nuda* as entities in scientific nomenclature provides for their consideration in post-original citations as something different from *nomina nova* (*sensu lato,* as defined in the preceding footnote), but it does not necessarily entail a revision of established policy concerning them. Having such recognition, however, the request that they become available only upon subsequent clear allocation by an author does not, as it would otherwise, involve all kinds of *nomina nova.*

As a minimal action, it is to be recommended that an Opinion on Article 18 clarify that *nomina nuda* are distinct entities that may attain the status of species names only upon clear allocation and adoption or acceptance of them as species names; and that the first worker to so treat them becomes their author. This recommendation is not to be construed as a precedent for like action regarding *nomina nova,* which even if proposed provisionally may acquire status if accompanied by adequate indication.

It should be made clear that I am not defending either the proposal of *nomina nuda* in modern literature or the adoption of them, where possible, by present-day students. It is to be hoped that modern taxonomists know correct usage of their nomenclatorial rules and would avoid such unfortunate circumstances. Errors do occur, however, and the present suggested ruling would outline what is to be done with them. Primarily, however, the proposed ruling is suggested as a means for treatment of *nomina nuda* and their subsequent citations in-past literature. It is for the earlier taxonomic literature that a guide for procedure is especially needed.

As an example of the effect of this ruling, suppose an author A inadvertently or otherwise puts into print a *nomen nudum, X-us albus.* A subsequent author (B), perhaps through acquaintance with A, knows what species A meant by *X-us albus,* and thus includes the name *X-us albus* in the synonymy of a species given another name, *Z-um nigrum.* Now suppose it is discovered later that the name *Z-um nigrum* is not available, and no other name for that species is available unless *X-us albus* is used. By the procedure under the present Rules, *X-us albus* B could be used, being available; by the interpretation recommended here it would not be available; author C could use the name for the species, but he would then be the accepted author of it; author C could also propose an entirely new name, and the *X-us albus* of authors A and B would have no effect upon it since it would be considered unavailable; furthermore if author D uses the name *X-us albus* for a completely different species, being unaware of the use by A and B, his name would not be invalidated by homonymity so far as A and B are concerned, by the recommended interpretation, although by the present Rules it would be invalidated.

IV

The preceding discussion has concerned only unacceptable and unavailable names that ordinarily can not be used as species names. Use of them as species names ordinarily would change their category to that of available names; and they can attain the status of available names only by that method.

All available names are here termed *nomina specialia.* They may (or may not) likewise be valid (*nomina valida*), depending upon whether or not they (1) can be identified within a reasonable probability (as of the date of the description) without reference to esoteric materials, (2) are not synonyms, and (3) are not homonyms. In considering any given *nomen speciale,* it is of prime importance to determine first its status as a *nomen clarum* or *nomen dubium. Nomina clara* are names whose diagnoses are originally adequate for identification and are likewise acceptable (as species names) under the terms of Article 25; such names are thus subject to all the Articles of the Code and Opinions thereon. They may be valid (*nomina valida*), homonyms (*homonomata*), or synonyms (*synonomata*); the latter may be *synonomata natura* or *synonomata regula,* according to whether zoological or nomenclatural reasons furnish ground for making the names synonyms. *Nomina dubia* differ from *nomina clara* only in having diagnoses originally inadequate for identification (see Opinion 126). Through the impossibility of definite specific allocation *nomina dubia* seldom can be (or can have) synonymic or homonymic names; they can never be *nomina valida* so long as they remain in their status of *nomina dubia.* They can be-

come valid only upon attaining the status of *nomina clara* (by procedures outlined in the following); and of course the attainment of that status can make them synonyms or homonyms instead of valid names, depending upon the circumstances. It may be recommended (1) that, unless it is the desire to fix the name and thus render it a *nomen clarum*, such names (*nomina dubia*) should be cited in conventional species-synonymies only with a question mark, and are thus to be placed with any species that seems a possible synonym; and (2) on the basis of the original description a name should be considered a *nomen dubium* rather than a *nomen clarum* only when originally the probability of correct association, after consideration of all generally available evidence (excluding esoteric information, as for instance, that furnished by a study of the type specimens) is reasonably low.

V

Except for *nomina clara* and *nomina dubia*, the proper taxonomic procedure in reference to the above categories is evident, at least for most cases. The procedure in reference to these two exceptions is somewhat complicated, but may be summarized:

I. *Nomina clara.* If a name has a diagnosis originally (*i.e.*, at the time of its proposal) adequate for identification, it is a *nomen clarum*. If a name is so considered, it can never be regarded as a *nomen dubium;* in other words, once a *nomen clarum*, always a *nomen clarum*, no matter how intricate the subdivision of the original species concept becomes. Under all circumstances the original author's name is retained. The name should always be allocated with some species, whether as a valid name, a synonym, or a homonym. Yet, it must be recognized that because of finer discrimination or for other reasons a diagnosis originally satisfactory may become inadequate for specific (or subspecific) determination.

A. Names based upon diagnoses which remain adequate.

1. Composite diagnoses. In case several species are clearly diagnosed with the name, the first reviser's restriction fixes the identity.
2. Single-species diagnoses.
   a. Type series of more than one species. As the diagnosis clearly refers to one species rather than to any other, the name can be applied only to the species diagnosed.
   b. Type series of one species. The object of legitimate taxonomy—a clear-cut, adequate diagnosis based upon a single species.

B. Names based upon diagnoses which later become inadequate for identification. All such names retain their original authorship, as of the original date of publication, regardless of subsequent history; and they suppress all homonyms, whether of the same species or not.

1. If holotype available and identifiable. Name to be restricted to the species to which it belongs.
2. If holotype (or other type material) available but unidentifiable. The name is to be restricted to any likely species; the allocation of the first reviser who places the name with an identifiable and likely species is to be accepted in preference to any others.
3. If there are several types, all alike, and identifiable. As in B1, above.
4. If there are several types, of different species. The name is to be restricted as of the first reviser to any identifiable specimen or material, or if none is identifiable, procedure as in B2, above.
5. If there are no types. As in B2, above.

II. *Nomina dubia.* If a name is accompanied by a diagnosis inadequate for identification even originally, it is a *nomen dubium*, and as such it is subject to very different rules from those that apply to *nomina clara*. They are to be treated practically like *nomina nuda*, save that they can suppress homonyms applied to species different from any of those to which the *nomina dubia* could reasonably apply, while homonyms based on what may be the same species are not suppressed unless they follow an earlier allocation. They can be restricted to a given species (becoming *nomina clara*) but then carry the name of the restrictor as senior author, that of the original author as junior (*e.g.*, Brown ex Green).[6] Moreover, *nomina dubia* may remain in that category as long as taxonomists care to avoid their definite allocation; they need never be rendered *nomina clara*. Fixation of them as *nomina clara* follows the same rules that govern availment of *nomina nuda*, within the following limits:

1. If a holotype is available, and identifiable: the name must be restricted, if at all, to the species of the holotype.

[6] This plan of double authorship follows Opinion 126 (p. 21). I rather doubt the wisdom of it. More in keeping with zoological practice would be simply to allow coauthorship of the usual type: Green and Brown, or Brown and Green, depending upon the decision of the International Commission. This question of multiple authorship is to a certain degree similar to that faced by botanists, whose procedure is to cite authors of any combination used. Fortunately zoologists have avoided that cumbersome procedure. Citation of multiple authors for *nomina clara* that have once been *nomina dubia* is not by any means the same, but it is possibly an entering wedge of complexity. The advisability of retaining only the original author's name (*e.g.*, Green) is seriously to be considered, in spite of the fact that Brown is the person who has fixed the name and made it of some use.

2. If a holotype (or other type material) is available but not identifiable: the name may be used for any species to which it may belong, and the allocation is to be that first proposed.
3. If there are several types, all alike and identifiable. As in II–1 above.
4. If there are several types, of different species: (a) the name is to be restricted as of the first reviser to any identifiable specimen or material; (b) or if none is identifiable, the name may be used for any species to which the diagnosis may belong, as of the first reviser.
5. If there are no types: as in II–4b, above.

Although *nomina dubia* can, as suggested here and as is generally practiced, remain forever without allocation, the proposal here put forth does jeopardize arrangements of any author (say A) who had neglected to allocate a *nomen dubium* that applies to a group of species all or some of which bear names of more recent date than the doubtful one. Another author (B) by reasonable allocation could invalidate one of A's names, and in fact would necessarily do so if the *nomina dubium* antedated all other names in the group and is otherwise available.

Recognition and allocation of *nomina dubia* is the most debatable of all procedures outlined here. Without question the entire matter of the status and treatment of them requires careful attention of taxonomists. In my suggestions I have merely followed and expanded in a seemingly logical manner the statement of Opinion 126, the only definite official discussion to date of this problem. Because of the lack of a clear ruling authors in the past have not always agreed upon allocation of *nomina dubia,* since many interpretations, all reasonable at least to some extent, are possible. Adoption of some procedure such as that outlined above would at least coordinate the actions of taxonomists, even though some might consider other procedures more useful for the greatest number of cases that may arise.

VI

The establishment of precise categories such as those suggested above may at first appear as unnecessary definitions of only academic importance. This is not so. No new concepts are suggested in these categories; they are merely concrete expressions of ideas long in common use by taxonomists but not well unified. Had the distinctions between them been made long ago, many of the difficulties encountered in the consideration of doubtful cases—either by individuals or by the commission—might well have been avoided. The existence of the concept of a *nomen clarum,* for instance, would have simplified markedly the discussion of genotypes in Opinion 65. The advantage of having categories in common use by taxonomists clearly stated, defined, limited and named, instead of hazily, incompletely or differently conceived or treated by them is beyond question a great one. While the definitions given are clearly unofficial, except for the acceptability of *nomina nuda* they reflect the opinions of the commission as revealed by study of the Code and Opinions. It is to be hoped that some official action along these lines may be forthcoming in the near future.

OBITUARY

HORACE CLARK RICHARDS

PROFESSOR RICHARDS died on May 20, 1945, in his seventy-eighth year. Since July 1, 1938, he had been emeritus professor of mathematical physics of the University of Pennsylvania. Except for two years, one spent at the Johns Hopkins University and the other at Bryn Mawr College, he had been associated with the university continuously since 1884, as undergraduate and graduate student, as Tyndale fellow in physics and as member of the staff of the department of physics. He was appointed professor of mathematical physics in 1914 and director of the Randal Morgan Laboratory in 1931.

Professor Richards's father was the first professor of architecture at the University of Pennsylvania. He designed the greenstone buildings erected on the present campus when it was established during the 1870's. The only brother of his father was an artist of distinction, whose canvases hang in America's leading galleries. Among the children of these two brothers there were three university professors, all in scientific fields, one at Columbia, one at Harvard, one at Pennsylvania, one of them a Nobel laureate.

Professor Richards was elected to the American Philosophical Society in 1907 and took an active part in its affairs until the end of his life. He contributed to its programs and served for many years as chairman of its library committee.

Early in his career Professor Richards saw that physics must reach out into ever-widening human associations. Accordingly, he took a leading part in the establishment of the Physics Club of Philadelphia in 1909. This club brings together teachers of physics in schools, colleges and universities, and physicists employed in industries of all kinds, and in laboratories connected with hospitals, schools of medicine,

dentistry and pharmacy. The club has enjoyed a continuous and vigorous existence. It is recognized as a regional chapter by the Association of Physics Teachers and as an associated society by the American Institute of Physics.

During 1918 Professor Richards served as visiting physicist at the National Bureau of Standards, working on problems of internal ballistics.

At the University of Pennsylvania he was the leader for thirty years in carrying on advanced instruction in physics, both at the graduate and the undergraduate levels. It was a heavy load, but he carried it joyously. His graduate courses on radiation and on the constitution of matter were masterpieces. He developed them early in the century and kept them up to date for more than two decades. His "Introduction to Mathematical Physics" attracted large numbers of students until the end of his teaching career. For a quarter of a century he always had students at work on optical problems. A series of studies of the optical properties of certain metallic elements and alloys was conducted under his supervision. His standards of scholarship were high and his influence upon his pupils was stimulating and lasting. All his associates developed a wholesome respect for his ideals and standards.

Professor Richards was a devoted alumnus of his college. He was keenly interested in the education of the undergraduates. He offered a series of courses for upper classmen that covered the whole field of classical physics. He gave one of these courses each semester with a thoroughness and a fidelity that won him the enduring gratitude of his pupils. He was not a lantern-slide and blackboard lecturer. His lectures were illustrated with demonstrations that required hours of preparation. He gave nature a chance to speak for herself and his pupils a chance to hear her voice.

Over a period of years Professor Richards encouraged teachers in the Philadelphia area to continue the study of physics and to work for graduate degrees in the field. The cumulative effect of his policy has been to create in the schools of the area a considerable group of teachers who hold advanced degrees in physics. To this group of people he was philosopher, guide and, above all, friend. They continued to visit him during the years of his retirement, and the hour of his funeral was set late in the afternoon to permit this group to be present.

In 1931 Professor Richards was invited to assume the directorship of the Randal Morgan Laboratory of Physics. It was not an easy assignment, for he was then in his sixty-fourth year, a depression reigned in the land, and the unrest and bewilderment abroad in the world expressed itself on his campus in cravings for "academic recognition" and in yearnings for "academic prestige." Professor Richards gave little heed to the stirrings around him, for he held with Russell Conwell that "acres of diamonds sparkle at every man's doorstep," and with S. Weir Mitchell that "the jewel Fame is found unsought along Duty's pathways." He was a modest, unassuming scholar and teacher and he had seen great honors worn with humility and dignity by some very near to him.

Professor Richards served as director of the laboratory for seven years, until he reached the age of retirement. During his last year of service pupils and other friends engaged John R. Pierce to paint his portrait. It was presented to the university at a testimonial dinner given during June, 1938. Professor Richards's response at this dinner brought editorial comment from Philadelphia's *Evening Public Ledger*. Under the title "Teacher's Secret" the *Ledger* said editorially:

> The guest of honor revealed with modest brevity his formula for successful teaching. "I always talked," he said, "as if somebody were listening." This is refreshing philosophy. . . . The teacher who talks "as if somebody were listening" is likely to have listeners and will be honored when his work is done.

This testimonial is submitted by pupils of Professor Richards.

R. DEWEES SUMMERS
WESTERN MARYLAND COLLEGE
THEODORE S. ROWLAND
NORTHEAST HIGH SCHOOL, PHILADELPHIA
E. A. ECKHARDT
GULF RESEARCH AND DEVELOPMENT COMPANY
R. C. DUNCAN
NAVAL ORDNANCE LABORATORY
THOMAS D. COPE
UNIVERSITY OF PENNSYLVANIA

DEATHS AND MEMORIALS

DR. DAVID LINN EDSALL, dean emeritus of the Harvard Medical School and of the Harvard School of Public Health, died on August 12 at the age of seventy years.

DR. HUGH CABOT, of the Mayo Clinic at Rochester, Minn., from 1930 to 1939 professor of surgery in the Graduate School of the University of Minnesota, died on August 14 at the age of seventy-three years.

DR. WILLIAM CRAMER, pathologist of the Barnard Free Skin and Cancer Hospital in St. Louis, died on August 10 at the age of sixty-seven years.

DR. ROBERT H. GODDARD, chief of Navy research on jet-propelled planes, died on August 10 at the age of sixty-two years. Dr. Goddard was formerly professor of physics and director of the laboratories of physics at Clark University.

DR. JOHN J. B. MORGAN, professor of psychology at Northwestern University, died on August 16 at the age of fifty-six years.

RICHARD BLAIR EARLE, known for his work in the development of synthetic rubber, died on August 13 at the age of sixty-nine years.

THE death is announced at the age of eighty-six years of Alexei Favorsky, of Leningrad, the organic chemist, well known for researches in the production of synthetic rubber.

THE Rochester, New York, Section of the American Chemical Society announces the inauguration during the coming year of an annual lecture to be established as a memorial to the late Dr. Harrison E. Howe to be known as the Harrison Howe Lectures. Dr. Howe was active in founding the Rochester Section and was one of its charter members. From 1921 until his death in 1942 editor of *Industrial and Engineering Chemistry,* he was nationally known as a lecturer. The lectureship is designed to provide discussion of topics of current importance in chemistry by outstanding authorities in the field. They will be presented each year before the Rochester Section as a part of its program. Provision is made for some variation in the form of the lectureship from year to year, the annual plans and selection of the speaker being entrusted to a lecture committee appointed each year. Instead of a single lecture, a series of two or three lectures on consecutive days may be decided upon if the subject is of special importance. An alternative form may be a symposium on a timely topic to permit presentation of papers by several speakers.

SCIENTIFIC EVENTS

THE BRITISH IRON AND STEEL RESEARCH ASSOCIATION

THE plan of the British steel industry of spending £120,000,000 on re-equipment will be backed by the expenditure of £400,000 a year on a new central research association.

This organization, according to a report in *The Times,* London, will receive up to £250,000 a year from the industry and its total revenue will be in the neighborhood of £400,000 a year. Dr. C. F. Goodeve, F.R.S., at present assistant controller for research and development of the Admiralty, has been made director. It will be known as the British Iron and Steel Research Association. To some extent, cooperative research has already been fostered by the work of the Research Council of the British Iron and Steel Federation, while the technical research workers in the industry have maintained mutual contact individually through the Iron and Steel Institute. The present plan, however, is to extend the field of cooperative research and exchange of information and also to encourage central research on matters of common interest.

Dr. Goodeve, before the war, was reader in physical chemistry at University College, London, and was for many years consultant and technical adviser to a number of industrial companies. At the Admiralty, as senior executive covering research and development for the Navy, he took a leading part both as scientist and as organizer of the work of other scientists in the technical discoveries which played a leading part in defeating magnetic mines, U-boats and other lesser but equally difficult weapons.

According to the present plan, research centers of the industry will work in connection with those of the universities. Teams are already working in Sheffield, Cambridge, Swansea, Glasgow, London, Birmingham and Newcastle. Full-scale development work will take place in or alongside works of member firms. The headquarters of the Research Association will be in London. The association will, on behalf of the industry, work with technical bodies studying raw materials, such as coal and refractories and design of plant used in making iron and steel. It will also bring together for common study of problems the users, such as the railways, shipbuilders and construction engineers, and the makers of iron and steel.

A PERMANENT STANDARDS ORGANIZATION

THE Executive Committee of the United Nations Standards Coordinating Committee, after an extensive survey of present conditions in the field of international standards and the rapidly changing events on the international scene, has come to the conclusion that the time is now ripe for setting up a permanent standards organization. The Executive Committee consists of the British Standards Institution, the Canadian Standards Association and the American Standards Association.

Invitations to attend the meeting have been sent out to the national standardizing bodies comprising the United Nations Standards Coordinating Committee. These are:

The Standards Association of Australia, Associacao Brasileira de Normas Tecnicas, The Canadian Standards

Association, The Chinese Standards Committee, The Association Française de Normalisation, The British Standards Institution, The New Zealand Standards Institute, The South African Standards Institution, The American Standards Association.

It is anticipated that representatives from practically all the countries will attend.

A full program of the detailed discussions to be undertaken at the meeting will be announced at an early date. In a general way, however, it can be said that the meeting will concern itself with the immediate problem of establishing the closest practical relations between the national standardizing bodies of the countries of the world; with the providing a forum through which these bodies can harmonize their activities internationally, and finally the meeting will deal with the major problem of integrating national standards and harmonizing them for the benefit of the total economy of the world.

THE MOENKOPI FORMATION OF NORTHERN ARIZONA

AT a meeting held in Flagstaff in the first week of July, an agreement was reached between the Museum of Vertebrate Paleontology of the University of California and the Museum of Northern Arizona for a joint cooperative study of the Moenkopi Formation of northern Arizona. In an announcement made by Dr. Harold S. Colton, director of the local museum, it is stated that the program of research will be conducted over a period of several years, and that the work done this summer is in the nature of reconnaissance. The meeting was attended by Dr. Sam Welles and Dr. Lyman Daugherty, of the Museum of Vertebrate Paleontology, and by Dr. Colton and E. D. McKee, of the Museum of Northern Arizona.

According to the announcement issued by the museum, "the Moenkopi formation is composed largely of red sandstone and shale with some limestone and gypsum. It covers the surface of the Plateau over wide areas, especially east and north of Flagstaff, where it lends color to the Painted Desert. Formed originally on a desert flood plain where streams deposited their sediments along the margins of an ancient sea that covered Nevada and parts of Utah, these rocks contain the bones of many peculiar and primitive animals, especially reptiles and amphibians. Inasmuch as this formation was formed at a critical period in the history of life and the earliest types of mammals appeared at that time, its study is believed to have considerable importance."

Excavation and examination of the skeletal remains will be done largely by Dr. Welles, who is an authority on the subject. Mr. McKee will attempt to work out the stratigraphy or interrelationships between the various rock types involved.

NEWS FROM ABROAD

DR. WM. RANDOLPH TAYLOR, of the University of Michigan, writes to SCIENCE that a letter received from Dr. Julienne Payen, student of the chemical constituents of the algae, indicates that difficulties in securing transportation to collecting areas interfered with her research during the war. She will now resume work at the Laboratoire de Cryptogamie, Museum d'Histoire Naturelle, Paris. A card has also been received from Dr. Ad. Davy de Virville, phycologist, indicating that he has become Directeur du Laboratoire des Travaux pratiques de Biologie végétale of the Sorbonne. His scientific collections escaped, though he had some personal losses due to the war.

A CARD signed by Professor Pierre Dangeard, Laboratoire de Botanique, Université de Bordeaux, France, dated April 12, reads in part: ". . . et j'ai le plaisir de vous informer que nos laboratoires et nos collections n'ont pas jusqu'ici souffert de la guerre. . . ."

A LETTER from Professor Roger Meslin, Caen, reads in part as follows:

Malheureusement notre vieille Université a été complétement anéantie par le feu et les bombes deux jours avant l'entrée des Alliés. En quelques heures des laboratoires, les collections zoologiques . . . , la riche bibliothèque universitaire ont detruits par l'incendie. Au Jardin des Plantes, les serres ont été egalement bombardées de même que la galerie des collections botaniques. Heureusement il n'y a pas en d'incendie et dans les decombes j'ai pu récoupérer la plus grande partie des herbiers algologiques, notamment celui de Lamouroux. Mais entrepôtes dans un local sans vitres, le herbiers ont beaucoup souffert des intempéries de l'hiver. Le laboratoire botanique, occupé par les allemands durant tout le mois de juin, a été pillé par eux, et le materiel volé ou demoli!

The collection of J. V. F. Lamouroux is one of the early nineteenth century algal collections most important for its reference value to phycologists, and its salvage a matter for sincere congratulation.

DR. J. H. F. UMBGROVE, Technische Hoogeschool, Delft, Holland, writes to Professor Marshall Kay, Columbia University: "My family has come through without any injury, though we have been through most trying times. . . . We have seen no English and American scientific magazines since May, 1940. Could you send as many reprints of geological, paleontological and geophysical articles as you and your colleagues can. I am really longing for them."

THE editor of *Chronica Botanica* reports that, ac-

cording to word received from reliable correspondents, the herbaria of Vienna are in good condition, with the exception of 2,500 bundles of herbarium specimens of flowering plants of the Natural History Museum, which had been stored and which were destroyed when the Russian army entered Ober-Höflein. The other collections of the Natural History Museum are safe in various places in the country. The collections and library of the Botanical Institute at the Rennweg are in good condition. The valuable library of the Zoologische-Botanische Gesellschaft, however, has been destroyed almost entirely.

SCIENTIFIC NOTES AND NEWS

THE Board of Directors of City Trusts of Philadelphia has announced the award of the John Scott Medal Fund jointly to Captain William N. Sullivan, Jr., Sanitary Corps, AUS, and Dr. Lyle D. Goodhue, who developed the aerosol bomb used for insect control purposes.. Captain Sullivan is at present stationed at the AAF Center at Orlando, Fla., where he is a member of the AAF Committee on Aerial Dispersal of Insecticides. Dr. Goodhue is senior chemist at the Beltsville, Maryland, Laboratory of the Bureau of Entomology and Plant Quarantine, U. S. Department of Agriculture.

DR. JAMES CREESE, since 1928 vice-president of the Stevens Institute of Technology at Hoboken, N. J., has been elected the sixth president of the Drexel Institute of Technology, Philadelphia.

DR. ALBERT RAY OLPIN, executive director of the research foundation of the Ohio State University, has been elected president of the University of Utah. He will take office on July 1, 1946.

DR. ELIOT BLACKWELDER, professor of geology and executive head of the department of geology at Stanford University, will retire at the end of this month. He is succeeded by Arville Irving Levorsen, consulting geologist of Tulsa, Oklahoma.

DR. CARL C. PFEIFFER, formerly chief pharmacologist of Parke, Davis and Company, was released from the Navy on August 15 to become head of the department of pharmacology, materia medica and therapeutics of the University of Illinois.

DR. VICTOR C. MYERS, director of the department of biochemistry of the School of Medicine of Western Reserve University, has been elected director of the newly established department of clinical biochemistry. It is planned to enlarge the department of biochemistry, of which Dr. Myers will continue as director until his successor has been chosen.

PROFESSOR JOHN R. WESKE, of the Case School of Applied Science at Cleveland, will join the department of aeronautical engineering of the Rensselaer Polytechnic Institute, Troy, N. Y.

DR. G. HARLOWE EVANS, of Huntingdon College, Montgomery, Ala., has been appointed to succeed the late F. S. Mortimer as head of the department of chemistry at Illinois Wesleyan University, Bloomington.

DR. C. L. HAMNER, assistant professor of pomology at Cornell University, has been appointed associate professor of horticulture at the Michigan State College, East Lansing.

DR. ELMER H. STOTZ has been named head of the new Division of Food Science and Technology that has been established at the New York State Experiment Station at Geneva by merging the Divisions of Bacteriology and Chemistry. The work of the group is to be guided by a committee consisting of members of the former divisions. This committee will consist of Dr. Stotz, who was head of the Division of Chemistry, *chairman;* Dr. George J. Hucker and Dr. Carl S. Pederson, professors of bacteriology; and Dr. Zoltan I. Kertesz, professor of chemistry.

DR. W. C. PIERCE, associate professor of chemistry at the University of Chicago, has been appointed head of the department of chemistry of Pomona College, Claremont, Calif.

DR. S. EDWARD SULKIN has been promoted to a professorship of bacteriology and immunology and has been made chairman of the department at the Southwestern Medical College, Dallas, Texas.

DR. H. F. STROHECKER, professor of zoology at Kenyon College, Gambier, Ohio, has been appointed professor of zoology at New Mexico Highlands University at Las Vegas, N. M.

DR. DONALD WAYNE TAYLOR, of the Radio Research Laboratory of Harvard University, has been appointed acting assistant professor of psychology and Thomas Welton Stanford fellow at Stanford University. He expects to assume his work there next January.

FURTHER appointments have been announced in the department of agriculture of the University of Minnesota: Dr. Clarence E. Mickel, acting chief of the Division of Entomology and Economic Zoology at the University of Minnesota since the retirement of Dr. William A. Riley on June 15, 1944, has been promoted to be chief of the division; Dr. Mykola H. Haydak has been promoted to an associate professorship of entomology and economic zoology. He will be in charge

of the beekeeping work of the division. He succeeds the late Dr. Maurice C. Tanquary; Dr. Laurence H. Cutkomp has been appointed research associate, and Dr. H. Y. Fan has been appointed research fellow, in the Division of Entomology and Economic Zoology. Both will assist in the research programs in insect physiology.

DR. J. C. C. MCKINSEY, assistant professor at Montana State College, has been appointed to an assistant professorship of mathematics at the University of Nevada.

DR. ROBERT S. BREED, head of the Division of Bacteriology of the New York State Agricultural Experiment Station at Geneva, at his own request has been relieved of administrative work as of August 1, after serving for thirty-two years as head of the division.

DR. EARL K. FISCHER, of the Research Laboratories of the Interchemical Corporation, has become head of the Division of Physical Chemistry of the Institute of Textile Technology, Charlottesville, Va.

DR. GARTH JOHNSON, formerly associated with the College of Medicine of the State University of Iowa and the Iowa State Department of Health, has been appointed director of microbiology at the Ortho Research Foundation, Linden, N. J. He will continue his work on the physiology and chemotherapy of *Trichomonas vaginalis* while directing similar investigations of associated organisms in the vaginal flora.

DR. RAYMOND C. OSBURN, emeritus professor of zoology and entomology of the Ohio State University, has accepted a research position at the Hancock Foundation, University of Southern California, Los Angeles, where he will be engaged for the coming year in the study of the marine Bryozoa of the Hancock Dredging Expeditions, 1932–42.

PROFESSOR ROBERT RAE, agricultural attaché at the British Embassy in Washington and agricultural adviser to the High Commissioner for the United Kingdom in Canada, will soon return to England. He will be succeeded by A. N. Duckham, who has been serving as director of the supply plans division of the Ministry of Food, having joined that department at the beginning of the war.

RESEARCH fellowships, given by the Imperial Chemical Industries Limited to certain British universities, include two fellowships at the University of Durham, each of the value of £600 and tenable in the first instance for three years, to Dr. W. E. Foster, for investigations into plant respiration and nutrition, and to Dr. G. A. Swan, for investigations in the structure of certain alkaloids.

W. L. MCATEE, technical director of the U. S. Fish and Wildlife Service, has deposited in the Manuscripts Division of the Library of Congress a handwriting collection that has been some forty years in the making. It is hoped that the material will be useful in the identification of unknown chirography, both printed and written. The collection may be profitably consulted also for biographical and historical details. It represents the following groups and approximate numbers of individuals: Botanists, 326; entomologists, 500; mammalogists, 140; ornithologists, 1,300; miscellaneous scientists, 423; the Biological Survey, 278; and the Wildlife Society, 136. For the first five of these classes, there remains duplicate material which Mr. McAtee will be glad to donate to any one interested. These residues will be allotted to persons or institutions heard from within a month of publication of this notice.

DR. H. C. BROWN, of Wayne University, has completed a series of five evening lectures at Purdue University which were attended by the staff and graduate students of the department of chemistry and others. Dr. Brown lectured on "Steric Strains in Organic Chemistry" and "High Vacuum Technique."

SIR ALEXANDER FLEMING, F.R.S., professor of bacteriology at St. Mary's Medical School, University of London, presented on July 18 an informal lecture on the development of penicillin before the staff and students of the Division of the Biological Sciences of the University of Chicago.

THE Porter fellowship has been awarded by the American Physiological Society to eighteen different individuals in the twenty years between 1921 and 1941. The annual stipend has been $1,200 given by Dr. W. T. Porter, of the Harvard Apparatus Company, after whom the fellowship is named. Dr. Porter has informed the society that in the future the stipend will amount to $2,400 annually and the society has announced that the award of this fellowship will be resumed in 1946.

FOUR research fellowships of the value of $1,500 each, established through funds provided by the Philco Corporation at Philadelphia, Pa., for study in the frozen food field, will become available at the Cornell University School of Nutrition in the autumn. The fellowships will provide one year of study each in frozen food economics, cookery, engineering and processing. Study in these fields already is under way. The fund provides "opportunity for the students to work into this field while carrying on graduate work."

UNDER the will of John Paine, of Troy, N. Y., bequests amounting to $500,000 have been made to Russell Sage College, and of $250,000 to the Rensselaer Polytechnic Institute.

THE new headquarters of the research laboratories

of the Navy Bureau of Ordnance will be constructed at White Oak, Montgomery County, Maryland, on ground covering 938 acres. This, with the Glenn L. Martin College of Engineering and Aeronautical Sciences of the University of Maryland, will provide for a center of research in the fields of physics and engineering.

GIFTS amounting to $25,000 have been made to the College of Medicine of the Ohio State University by Franz T. Stone, Columbus, in honor of the ninetieth birthday of his father, chairman emeritus of the Board of Trustees. One gift, in the amount of $20,000, goes to establish the Julius F. Stone Fund for Medical Research, the income to be used for research in the field of physical medicine. This fund will be administered by a committee including the dean of the College of Medicine, the chairman of the Department of Medicine and the dean of the Graduate School. The second gift of $5,000 is made to found a Julius F. Stone medical fellowship, for research in the division of physical medicine. The first recipient of this fellowship is Dr. William G. Myers, research associate in the department of bacteriology.

THE School of Medicine of the University of Utah is the recipient of a recent grant of $92,000 from the U. S. Public Health Service for research in muscular dystrophy and related degenerative disorders. A considerable number of cases are available in Utah and near-by states for study, and it is explained that church records and other local factors may throw considerable light on genetic aspects of the research. Dr. M. M. Wintrobe, professor of medicine, is director of the study and an advisory committee of known scientists will be appointed.

CONSTRUCTION will start next spring on a new Structural Research Laboratory at the University of Washington, Seattle. The laboratory will contain four new testing machines, one of which is now being designed and constructed by the Baldwin Southwark Division of the Baldwin Locomotive Works, Philadelphia. Representing the latest word in testing technique and incorporating important features not found in any existing machine, it will have a capacity of 2½ million pounds compression and 2 million pounds in tension. It will accommodate specimens 10 feet wide and as a transverse tester it will accommodate structural assemblies in excess of 80 feet in length and can subject them to a total transverse load of 2½ million pounds—a combination of length and loading much beyond the capacity of any equipment yet built.

APPLICATIONS for grants from the Elizabeth Thompson Science Fund should be made to the secretary, Dr. Jeffries Wyman, Woods Hole Oceanographic Institution, Woods Hole, Mass.

AN agreement has been made by the City of New York and the Columbia-Presbyterian Medical Center for the construction and joint operation of a hospital and health center for the study and treatment of tropical and communicable diseases, to be constructed at a cost of $5,390,000. According to the agreement, it will be built on land conveyed without cost to the city by Columbia University and the Presbyterian Hospital. The site extends 800 feet along the west side of Riverside Drive between 165th and 168th Streets. Two structures are planned—a three-hundred bed hospital and a research building. The latter will house the diagnostic laboratory of the Department of Health, a research center for the Public Health Research Institute and a specialized branch conducted by the Medical School of Columbia University. The capital budget counts on Federal funds for the entire $2,700,000 required for the hospital and for half the $2,690,000 allocated for the research center.

THE establishment has been announced of the Research Foundation of the Oklahoma Agricultural and Mechanical College, following the passage of a bill by the Oklahoma Legislature, authorizing expansion of the research activities of the college. The purpose of the Research Foundation is to coordinate research at the college and to support research in fields lacking formal research problems. The work is under the direction of Drs. K. Starr Chester, director of research; Dr. Otto M. Smith, director of negotiation, and Schiller Scroggs, director of administration. The program at present includes projects in the fields of chemistry, veterinary medicine and wildlife conservation.

A COMPREHENSIVE study of hill culture has been started at the West Virginia Agricultural Experiment Station with the objective of increasing the income of several thousand small hill farmers. Experiments include the production and marketing of holly, native nuts and nut meats, wild fruits, maple sugar, etc. The project is being financed from a grant by the Sears Roebuck Foundation.

THE Nutrition Foundation, New York City, has issued a pamphlet giving a list of the grants that have been made. These are grouped under the following headings: "Human Requirements of Individual Nutrients"; "The Origins and Functions of Individual Nutrients"; "Maternal and Infant Nutrition"; "Public Health Problems in Nutrition"; "Education and Professional Training" and "Nutrition Studies Related Directly to the War Emergency."

THE Michigan College of Mining and Technology at Houghton has established a Timber Products and Forest Industries Institute to serve and foster the wood industries of Northern Michigan. Its staff will

conduct research, provide a center of information, hold schools of instruction and demonstration, and organize and direct a practical course in woods industries of two twelve-week terms and one nine-week term.

ERRATUM: Page 148, 2d column, article by Dr. Gregory Shwartzman; the thirteenth line down on the right-hand column reads: "experiments which were all carried out in meat-inferior broth." This should have been "meat infusion broth."

SPECIAL ARTICLES

REVERSIBLE PARALYSIS OF MOTOR FUNCTION IN RATS FROM THE CHRONIC ADMINISTRATION OF DITHIOBIURET

DURING the course of investigations on the antithyroid activity of compounds related to thiourea it was observed that small concentrations of dithiobiuret (NH_2–C(=S)–NH–C(=S)–NH_2) were lethal within seven days.[1] It was soon apparent that death was preceded by paralysis and was presumably due to involvement of the respiratory muscles. This phenomenon was found to be readily reproducible and the effective dosage of dithiobiuret[2] proved to be quite critical. When administered in the drinking water in a concentration of 0.002 per cent. no effect was observed for 2 to 4 days and the animals continued to gain weight; then weakness of the hind quarters became apparent. Soon thereafter the muscles of the entire body with the exception of the muscles of respiration and those of the head and neck became completely paralyzed and the animals lay on their sides breathing with difficulty. If continued in this dosage the compound was uniformly lethal at the end of 5 to 6 days, but if reduced to 0.001 per cent. the animals would survive. By appropriate adjustments in the concentration of the drug between 0.001 per cent. and 0.002 per cent. the animals could readily be maintained in a state of profound muscular paresis for many weeks. With the onset of muscular weakness food and water intakes decreased and the animals began to lose weight; food and water were procured at the expense of great effort. The only constant associated finding was an incrustation about the eyes containing the red porphyrin pigment from the Harderian glands. There later ensued progressive weight loss, muscular wasting and contractures of the paralyzed muscles. The condition was consistently reversible, for despite the presence of extensive paralysis complete recovery followed a few days after withdrawal of the drug; even when atrophy and contractures had become manifest good recovery of muscle function followed within a week or two.

A single subcutaneous or intraperitoneal injection of relatively huge doses of this substance was well tolerated and failed to cause paralysis. A dose of 20 to 50 mg was necessary to kill an adult rat from a single injection, whereas fatal paralysis ensued in 5 days from the daily administration of less than 0.5 mg either in the drinking water or by subcutaneous injection.

The paralysis seemed not to be due to a disturbance of the muscles themselves, of the peripheral nerve or of the myoneural junction, for faradic stimulation of the motor nerves elicited muscular contraction. Nor did there seem to be any disturbance of pain sensation as judged by the responses to painful stimuli in partially paralyzed animals.

The brains, spinal cords and roots, spinal ganglia, sciatic nerves and muscles of animals which had been kept paralyzed for two to three weeks and of control animals were removed and sections were stained for cells, axis cylinders, myelin and fat. Microscopic examination disclosed no visible structural damage, a finding which is in keeping with the reversible nature of the disturbance. Determinations on the content of acetylcholine and of choline esterase in the brains of paralyzed rats yielded normal values.[3]

Attempts to elucidate the phenomenon by the administration of drugs which affect the nervous system and by a search for a remedial agent were unsuccessful. Strychnine failed to induce convulsions in paralyzed animals and double the usual lethal dose was required to produce death, which was accompanied by a slight flexure of the head upon the thorax and cessation of respiration. Pilocarpine and prostigmine did not improve muscular function in doses up to the lethal level. Atropine, epinephrine and ephedrin were likewise ineffective. Large doses of a crude liver extract, thiamin, nicotinic acid, vitamin A, biotin, brewer's yeast or biuret failed to prevent paralysis and caused no improvement when administered to paralyzed animals.

These observations appear to be of interest in that they probably point to the existence of a new mechanism which is essential to the transmission of nervous impulses within the central nervous system. They

[1] E. B. Astwood, *Jour. Pharm. and Exper. Therap.*, 78: 79, 1943.

[2] This compound was made available through the kindness of Dr. R. O. Roblin, of the American Cyanamid Co., Stamford, Conn.

[3] We are indebted to Mr. J. C. Seed for the acetylcholine determinations and to Miss Mary Root for the assays of cholinesterase.

indicate that the chronic administration of this simple chemical compound interferes with a hitherto unrecognized process, perhaps by making unavailable a component of some enzyme system.

E. B. ASTWOOD
A. M. HUGHES
M. LUBIN
W. P. VANDERLAAN
R. D. ADAMS

DEPARTMENTS OF PHARMACOLOGY,
MEDICINE AND NEUROLOGY,
HARVARD MEDICAL SCHOOL

THE MECHANISM OF ACTION OF ALLOXAN ON BLOOD SUGAR

THE intravenous administration of alloxan induces a triphasic modification of the blood sugar level: (1) hyperglycemia; (2) hypoglycemia; (3) hyperglycemia. We have studied these phenomena in several species, particularly the chlorolosed dog (100 mg of alloxan/kg of body weight) and the toad *Bufo arenarum* Hensel (200 mg/kg).

The initial hyperglycemia did not appear in hepatectomized dogs or toads nor in eviscerated dogs. It was observed in adrenalectomized animals (5 dogs and 6 toads) and in 3 dogs with previous section of the splanchnic nerves (major and minor) so that it can not be attributed to either adrenaline or cortical hormones. It was also observed in recently hypophysectomized toads. If injected in the portal vein, alloxan produces a higher initial hyperglycemia (5 dogs) than if injected in the saphenous vein (8 dogs).

The secondary hypoglycemia is not due to liberation of insulin by the β cells of the islets undergoing destruction. Nine dogs totally depancreatized half an hour before injection of alloxan, showed a marked hypoglycemia beginning 1, 2, 2, 2, 2, 3, 3, 4 and 5 hours after injection; the blood sugar level reaching in 7 cases to 50 and 24 mg per 100 cc. Six of these dogs showed initial hyperglycemia. Pancreatectomized controls with no alloxan only in a few cases showed slight and brief diminution of the blood sugar level half an hour after the operation, followed by a gradual and steady increase from 2 to 6 hours after the operation, reaching at that time 0.149 and 0.180 g per 100 cc of blood.

In 7 dogs depancreatized 24 to 48 hours previous to the injection of alloxan there was no hypoglycemia: on the contrary, the blood sugar level was slightly increased. In only one case was there a moderate decrease (from 0.217 to 0.134 g per cent. between 5 to 6 hours after the injection).

In pancreatectomized toads, alloxan injected immediately after the operation either prevents or decreases the diabetic hyperglycemia during the next 24 hours. If injected 24 hours after pancreatectomy, the existing diabetic hyperglycemia decreases as shown by the blood samples 24 hours after injection. Alloxan also notably decreased the diabetogenic action of the *pars distalis* of the hypophysis when subcutaneously injected to the hypophysectomized and depancreatectomized toad.

The capacity of the pancreas to secrete insulin was investigated by grafting in the neck through vascular anastomosis the duodeno-pancreas of dogs to dogs rendered diabetic through pancreatectomy performed 24 hours before. Normal pancreas decreases the blood sugar to normal level within 3 to 5 hours. Pancreas from dogs injected with alloxan 24 hours (6 dogs) or 48 hours (2 dogs) before extraction and grafting did not secrete insulin in 4 cases, the secretion was very reduced in 3 cases and only in 1 was the secretion normal. It is interesting to note that the pancreas was taken in some cases from animals that were still hypoglycemic. The β cells of the islets showed lesions in all cases (Dr. Di Pietro).

The final rise of the blood sugar reaches sometimes (rats, rabbits and dogs) higher values to those usually observed after pancreatectomy. Values of 0.700 and 1.00 g per 100 cc of blood have been observed. Possibly the liver plays some part in this phenomenon.

Therefore: (1) The liver is essential for the initial hyperglycemia produced by alloxan. Hyperglycemia is observed in adrenalectomized animals and those with section of the splanchnic nerves. It must be attributed principally to a direct action of alloxan on the liver. (2) The secondary hypoglycemia is not due to liberation of insulin, but to an extrapancreatic effect: probably lack of glucose production by the liver. The liver of the animal already in a diabetic condition is generally insensible to this action of alloxan. (3) The final hyperglycemia is mainly due to the destruction of the β cells of the islets of Langerhans, and becomes permanent if the animal survives. (4) The liver plays an important role during the 3 phases of modification of blood sugar level.

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THE PRODUCTION OF ANTI-PENICILLINASE IMMUNE SERA

IT is well known that the injection of an antigen (precipitinogen) parenterally in animals stimulates the production of antibodies (precipitins). In order that a substance may be precipitinogenic, it apparently must contain a soluble protein.[1]

[1] F. P. Gay and Associates, "Agents of Disease and Host Resistance," Charles C Thomas, Baltimore, Md., 1935.

The penicillinase prepared in our laboratories[2] for penicillin inactivation consists of about 5 per cent. protein. Since this preparation was not tolerated well by rabbits, it became necessary to prepare specially purified penicillinase[3] for this purpose. It was believed that immune sera could be prepared using this latter preparation of penicillinase (containing about 8 per cent. protein) as the antigen.

METHODS

Preliminary experiments with rabbits were conducted to determine the amount of antigen to be injected, the number of injections and the interval between the injections, which was necessary to induce the formation of the immune bodies. The diluted antigen was injected slowly in small, graded doses in order to prevent sensitization and associated reactions. The intravenous route was used since purified penicillinase was not very toxic for rabbits and there was little danger of eliciting anaphylactic responses by repeated injections. The injections of antigen were made in the marginal vein of the right ear.

Two series of injections were found to give reliable results. 1.0 mgm of protein per kilogram of body weight was given five times weekly for two weeks and then 2.0 mgm of protein per kilogram of body weight five times during the third week, bleeding was made daily from the marginal vein of the left ear. Prior to bleeding the animals were fasted overnight so as to avoid serum opalescence usually due to an increase in the lipoidal elements of the serum from digesting food.

When the serum from this preliminary bleeding was found to be sufficiently high in antibody titer (usually five to ten days after the last injection) the animal was anesthetized and bled from the heart. The blood was allowed to clot, then centrifuged and the serum pipetted from the top. The serum was then Seitz filtered and stored in the refrigerator to "age" for at least one week before subsequent testing for titer. This was done so as to remove any substances which may precipitate spontaneously. When any such non-specific materials precipitated, the serum was again Seitz filtered.

The precipitin test was performed using the so-called serum dilution method as described by Culbertson.[4] The tubes were incubated at 37° C for two hours to obtain practically complete precipitation[5] and then stored in the refrigerator overnight. The following were the maximum titers obtained after a three-week injection series (see Table 1).

TABLE 1

TITER OF IMMUNE SERUM COMPARED WITH NORMAL SERUM

	Serum dilution	Penicillinase added		Precipitins
		mgm	units	
Immune serum	Str.	0.25	100	++++
	1:2			++++
	1:4			++++
	1:8			+++
	1:16			++
	1:32			++
	1:64			+
	1:128			±
	1:256			0
	1:512			
Normal serum	Str.	0.25	100	0
	1:2			0
	1:4			0
	1:8			0
	1:16			0
	1:32			0
	1:64			0
	1:128			0
	1:256			0
	1:512			

It will be noted that a positive precipitin test was obtained in a serum dilution as high as 1:256, whereas the normal serum was negative in all dilutions. Experiments are now being conducted to further increase the antibody content of the immune sera. Further work will be initiated to obtain the gamma globulin fraction by purification of the serum, which presumably contains practically all the antibody activity.[6]

There are three main routes for the inactivation or excretion of penicillin[7] in the body, the gastro-intestinal tract, the renal system, and oxidation, reduction or conjugation.[8] The inactivation in the gastro-intestinal tract is caused to a considerable extent by the penicillinase produced by certain intestinal bacteria.

It was therefore believed that the combination of this immune serum globulin fraction with penicillin will not only delay the action of penicillin but should also protect it from inactivation by penicillinase producing organisms in the animal body. This work is now under investigation and will be reported at a later date.

SUMMARY

It has been found that an anti-penicillinase immune serum can be produced in rabbits by the use of purified penicillinase as the antigen.

Anti-penicillinase serum is suggested as protection for penicillin from inactivation by penicillinase.

Acknowledgments: The authors wish to express their sincere appreciation to E. B. McQuarrie for his

[2] E. B. McQuarrie, A. J. Liebmann, R. G. Kluener and A. T. Venosa, *Archiv. Biochem.*, 5: 307, 1944.

[3] Supplied by Mr. E. B. McQuarrie and associates of the Biochemical Division of Schenley Research Institute.

[4] J. T. Culbertson, *Jour. Immunol.*, 23: 439, 1932.

[5] J. G. Baier, *Proc. Soc. Exp. Biol. and Med.*, 27: 421, 1929.

[6] J. F. Enders, *Jour. Chem. Invest.*, 23: 510, 1944.

[7] K. H. Beyer, L. Peters, R. Woodward, W. F. Verwey, *Jour. Pharmacol. and Exp. Ther.*, 82: 310, 1944.

[8] D. Perlstein, H. E. Wright and A. J. Liebmann, SCIENCE, 101: 562, 1945.

valuable suggestions and to I. Dorrell and D. Klingelhoffer for their technical assistance.

DAVID PERLSTEIN
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BLOOD LEVELS AND URINARY EXCRETION IN PEANUT OIL, BEESWAX AND PENICILLIN MIXTURE

CLINICALLY, penicillin has proven to be a highly effective drug, yet its application in non-hospitalized patients is a difficult problem because of its rapid utilization and excretion from the body. This has necessitated repeated injections at frequent intervals; a procedure that more or less disrupts both patients' and doctors' daily schedules. Recently several methods of prolonging the action of this drug have been described. Of these, the most promising is the use of a penicillin-peanut oil and four per cent. beeswax mixture, as described by Romansky and Rittman[1] and confirmed by Zinnamon and Seeberg.[2] The prolongation is achieved by delaying the absorption of penicillin from the area injected.

Recently, an opportunity to use this mixture was afforded this department, and the data obtained pertaining to the excretion of this drug form the basis of this report. The clinical results obtained were good, but will not be considered here, as a more complete report of these findings has been published by the U. S. Public Health Service.

TECHNIQUE

The material used was a mixture of peanut oil and four percent. white beeswax containing 100,000 units of calcium penicillin per cc.[3]

Urine and blood were assayed by the serial dilution "turbidimetric" method of M. H. Dawson and G. L. Hobby.[4] The number of Oxford units were determined by comparing bacterial inhibition of serial dilutions of the fluid to be tested and sets of penicillin solutions of known potency determined by assay against a reference standard of penicillin calcium obtained from the Food and Drug Administration of the Federal Security Agency. The bacteria used were the Oxford strain of hemolytic *Staphylococcus aureus*. This is not the method best suited for the determination of minute amounts of penicillin, as this strain is inhibited by 0.1 units per cc. In our hands, however, it is rapid, practical, and gives easily reproducible results.

The drug was administered intramuscularly in a single dose of 200,000 units. Blood and urine specimens were assayed at the intervals shown in the following table and interval and total excretion values were calculated. A total of 23 patients were treated. The values shown represent the average amounts of penicillin found for each interval. The minimum and maximum amounts are also given.

TABLE 1

PENICILLIN EXCRETION FROM BEESWAX AND PEANUT OIL MIXTURE

Time interval hrs.	Blood levels (Units per cc) Low	High	Average	Urine levels (Units per cc) Low	High	Average	Urine levels (Total units excreted) Low	High	Average
2	.225	.45	.343	45.	135.	109.5	3,600	18,225	10,660
4	.225	.45	.235	33.7	135.	83.6	3,375	18,900	8,790
6	0	.23	.152	22.5	135.	75.3	0	17,100	7,672
8	0	.225	.020	22.5	90.	61.4	1,854	14,400	6,884
10				22.5	135.	69.1	2,812	11,250	5,838
12				22.5	135.	54.3	2,250	13,500	5,333
14				22.5	90.	48.4	1,800	13,500	5,323
16				0	67.5	38.1	0	14,087	4,495
18				0	90.	35.1	0	7,414	3,104
20				0	90.	33.4	0	6,000	2,495
22				0	67.5	23.5	0	4,275	2,233
24				0	67.5	20.0	0	5,063	1,786
24 to 36				0	45.	9.5	0	15,750	4,866
36 to 48				0	11.5	1.7	0	5,175	640
Totals ..							15,691	164,639	70,119

In short, our findings confirmed the prolongation of demonstrable blood levels as reported by Romansky and Zinnamon. An additional fact, however, which was noted and considered of more import than has previously been accorded it, was the even more prolonged presence and concentration of penicillin in the urine. The implications of this will be considered later.

DISCUSSION

It will be noted that penicillin could be detected in significant amounts in the blood for an average of over five hours. Since all reports[5, 6, 7] thus far agree that the water or saline solutions of penicillin are excreted in 3 to 4 hours, this is a definite prolonga-

[1] Romansky and Rittman, *Bull. U. S. Army Med. Dept.*, No. 81, p. 43, October, 1944.

[2] Zinnamon and Seeberg, *Venereal Disease Information*, 26: 2, 27, February, 1945.

[3] Prepared and furnished by Squibb in cooperation with the U. S. Public Health Service.

[4] Dawson and Hobby, personal communication.

tion. Actually, the degree of prolongation may be greater as the assay methods used by the above investigators gave values as low as .01 unit per cc, whereas the method in this report had a minimum reading of .1 unit per cc.

TABLE 2

SHOWING DEGREE TO WHICH PENICILLIN IS CONCENTRATED BY KIDNEY

Case	Interval Hours	Blood units/cc	Urine units/cc	Concentration
No. 15	2	.225	135	?
	4	.225	135	600
	6		135	600
No. 17	2	.45	90	
	4	.225	90	200
	6	.225	90	400
	8		37.5	150
No. 20	2	.225	90	
	4	.225	135	600
	6	.225	45	200
	8		37.5	150

During this work it became apparent that penicillin was consistently present in the urine in much greater concentration than in the blood stream and that it may be detected much longer. The rate of excretion dropped rather rapidly in the first 6 to 8 hours and then remained fairly constant for 8 to 16 hours despite the low or absent blood levels. The unit excretion per interval of time was for the most part independent of the urinary volume.

It has been suggested that penicillin is excreted from the tubules of the kidney in addition to filtration through the glomeruli. Concentration values were calculated on the basis of urinary concentration in units per cc divided by the blood level at the end of the previous two-hour period. Sample concentrations appear in Table 2 and varied from 100 to 600 times. As the kidney excretes non-threshold substances creatinine and sulfates in concentrations of 75 to 90 times, respectively, the markedly high values found with penicillin can be explained only by renal tubular excretion. Dawson *et al.*[8, 9] have shown that blood serum and whole blood do not inhibit penicillin, therefore the persistence of high concentrations of penicillin in urine long after demonstrable blood levels are absent can be explained by the remarkable power of the kidney to concentrate and excrete it. Another explanation might be storage in the tissues, which would tend to make the blood levels lower and prolong the excretion in the urine.

The total amount of penicillin excreted was relatively constant around 68,000 units, which represents 34 per cent. of the original dose given. This rather low total excretion when compared to values of 50 to 60 per cent. excretion after injection of penicillin in water or saline can be explained by its destruction in the body due to its heat lability or to some other factor in which the time element plays a part. Rammelkamp and Keefer[7] have shown that while the liver excretes penicillin in bile, the total excretion is probably small.

The prolonged presence of penicillin in the urine, however, does suggest the possibility of penicillin being clinically available in the body long after our present concepts have led us to believe. It is true that the levels are minute, but for sensitive organisms and in the presence of leucocytes and antibodies they may prove sufficient.

CONCLUSIONS

(1) Penicillin in combination with peanut oil and beeswax is detectable in the blood stream for longer periods of time following its intramuscular injection than when a water or saline suspension is used.

(2) Penicillin is present in the urine in greater concentration and for much longer intervals than in the blood stream, the concentration being 100 to 600 times. This may prove to be of clinical significance and of value in studying renal physiology.[10]

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IMMUNIZATION AGAINST MALARIA: VACCINATION OF DUCKS WITH KILLED PARASITES INCORPORATED WITH ADJUVANTS[1, 2]

IN his recent review of immunity in malaria, Coggeshall[3] came to the conclusion that "the acquisition of immunity following the inoculation of killed malarial organisms is only demonstrable under exceptional conditions." Jacobs[4] published evidence indicating that partial immunity against *P. lophurae* in the duck may be obtained by injecting killed parasites in combination with staphylococcus toxoid. In his experiments the immunized ducks were challenged three days after the fifth injection of vaccine and

[5] Herrel, Nichol, Heilman, *Jour. Am. Med. Asn.*, 125: 15, August 12, 1944.
[6] Cooke and Golding, *Jour. Am. Med. Asn.*, 127: 80, January 13, 1945.
[7] Rammelkamp and Keefer, *Jour. Clin. Invest.*, 22: 425, May, 1943.
[8] Rammelkamp and Bradley, *Proc. Soc. Exp. Biol. and Med.*, 53: 30, May, 1943.
[9] Dawson, Hobby, Meyer and Chaffee, *An. Int. Med.*, November, 1943.

[10] It is desired to acknowledge appreciation to Sergeant Hugh Woosley, who performed the laboratory work herein reported.
[1] Manuscript completed December, 1943.
[2] This study was aided by a grant from the John and Mary R. Markle Foundation.
[3] L. T. Coggeshall, *Medicine*, 22: 87–102, 1943.
[4] H. R. Jacobs, *Am. Jour. Trop. Med.*, 23: 597–606, 1943.

some of the animals showed very little if any protection. Recent experiments,[5,6,7] have shown that antibody production against horse serum, diphtheria toxoid, bact. typhosum and other antigens can be enhanced and sustained for a remarkably long time by combining antigens with a lanolin-like substance and paraffin oil, with or without killed tubercle bacilli.

In the present work the same adjuvants were applied to immunization against malarial infection with *P. lophurae* in the two-months-old White Pekin duck. Heavily parasitized duck red blood cells (approximately 100 parasites per 100 red blood cells) were suspended in salt solution containing 0.1 per cent. formaldehyde, kept at 4° C over night and washed three times with saline. A heavy suspension was mixed with a lanolin-like substance (Falba[8]) and paraffin oil with or without killed tubercle bacilli (Table 1). The proportion was one part red blood cell suspension, one part Falba, and one and one half parts paraffin oil. The vaccine was injected in divided doses into the subcutaneous tissue and muscles over the lateral parts of chest wall. The intervals between injections were approximately one month. Formalin-treated parasitized red blood cells produced no infection when injected intravenously in young ducks. Normal ducks of the same age and kept under the same conditions as the immunized ducks served as controls.

TABLE 1

DOSAGE OF KILLED P. LOPHURAE AND KILLED AND DRIED TUBERCLE BACILLI

Injection No.	Experiment 1		Experiment 2	
	Parasites billions	Tubercle bacilli mg	Parasites billions	Tubercle bacilli mg
1	5	1.0	15	1.7
2	21	0.2	12	0.1
3	11	0	11	0

TABLE 2

NUMBER OF PARASITES PER 100 RED BLOOD CELLS

Group	Duck No.	Day after infection 1	2	3	4	5	6	7	8	9	10	11	12
Experiment 1													
Immunized	27	.2	.6	.8	1	5	8	Dead	(1)				
	34	0	.4	.6	.6	.2	.2	0	0	0	0	0	0(2)
	28	0	.6	.2	0	0	0	0	0	0	0	0	0(2)
	36	0	.4	0	.2	.2	0	0	0	0	0	0	0(2)
Control	35	.2	.2	1	3	13	30	63	83	157	Dead		
	31	.4	0	2	3	9	10	24	58	93	77	Dead	
	37	0	.6	.4	1	6	4	16	42	80	83	49	8(3)
	38	0	.2	1	2	3	3	10	30	30	54	28	6(4)
Experiment 2													
Immunized	43	.2	.2	2	4	0	.4	0	0	0	0	0	0(6)
	41	.4	1	3	–	1	.6	.4	0	0	0	–	0(5)
	40	.2	0	0	0	0	0	0	0	0	0	0	0(6)
	39	0	0	0	0	0	0	0	0	0	0	0	0(6)
Control	44	.4	.2	1	5	8	21	59	102	128	98	Dead	
	47	.2	0	.8	4	4	21	34	76	88	108	Dead	
	48	0	.4	1	.8	2	2	2	5	16	22	37	22(7)
	45	0	0	.8	1	.2	.6	.2	1	5	7	15	6(8)

– Not done.
(1) 350 ml fluid in the peritoneal cavity.
(2) No parasites found through 23rd day.
(3) Became 0 on 18th day.
(4) Became 0 on 13th day.
(5) Duck 41 received only the first and second injections of vaccine and was infected 4 weeks later. No parasites found through 41st day.
(6) No parasites found through 13th day.
(7) Became 0 on 20th day.
(8) Became 0 on 20th day.

Both immunized and control animals received approximately one billion parasites by intravenous injection about one month after the last immunizing injection. All ducks were examined for parasitemia by counting parasites in at least 500 red blood cells daily for thirty days after the infecting dose.

Two experiments were carried out using four immunized and four control ducks in each. In the first experiment, as Table 2 shows, three immunized ducks had parasites for only a few days and the counts were less than one parasite per 100 red blood cells. The fourth duck (No. 27) died with 350 ml turbid fluid in the peritoneal cavity and a layer of fibrin covering the liver. In this duck the number of para-

[5] (a) J. Freund and K. McDermott, *Proc. Soc. Exp. Biol. and Med.*, 49: 548–553, 1942. (b) J. Freund and M. V. Bonanto, *Jour. Immunol.*, 48: 325–334, 1944.

[6] (a) K. Landsteiner and M. W. Chase, *Proc. Soc. Exp. Biol. and Med.*, 49: 688–690, 1942. (b) M. W. Chase, *Proc. Soc. Exp. Biol. and Med.*, 52: 238–240, 1943.

[7] (a) L. M. Kopeloff, S. E. Barrera and N. Kopeloff, *Am. Jour. Psychiat.*, 98: 881–902, 1942. (b) L. M. Kopeloff and N. Kopeloff, Federation Proceedings (Am. Soc. for Exp. Biol.), 2, No. 1, 99, 1943.

[8] Manufactured by Pfaltz and Bauer, Inc., New York City.

sites reached 7.6 per 100 red blood cells. Of the control ducks three showed more than 80 parasites per 100 red blood cells and the other 54 parasites per 100 red blood cells. Two of these ducks died. In the second experiment the protection was almost as great as in experiment 1. None of the control ducks which died showed on autopsy any gross changes indicating death from any cause other than malaria.

Discussion

Although we have no experiments with malaria on this point, other experiments[5] in which the adjuvants mentioned above were used combined with various antigens suggest that one single injection of malarial parasites plus adjuvants may be sufficient to produce the results described above and that the protection may last for a very long time. It is also possible that similar results could be obtained without tubercle bacilli or that timothy-grass bacilli can be substituted for tubercle bacilli.[5b]

In the interpretation of our results it may be mentioned that the absence of parasites in 500 red blood cells does not indicate that there are no parasites in the blood or in the organs. Thus in these experiments the actual immunity may be less than the apparent disappearance of the parasites from the blood of the immunized ducks suggests.

Summary

When ducks are injected with formalin-inactivated *P. lophurae* in combination with a lanolin-like substance, paraffin oil and killed tubercle bacilli, they develop considerable resistance to subsequent infection with *P. lophurae*.

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IMMUNIZATION OF RHESUS MONKEYS AGAINST MALARIAL INFECTION (P. KNOWLESI) WITH KILLED PARASITES AND ADJUVANTS[1,2]

Eaton and Coggeshall[3] reported that numerous injections of large doses of *P. knowlesi* parasites killed by formalin or other means do not produce sufficient immunity in Rhesus monkeys to protect them against lethal infection with the same strain of parasite. Coggeshall[4] found, however, that immunity did develop in monkeys with chronic infections (carriers) since they were resistant to newly introduced parasites (*P. knowlesi*) of the same strain. Moreover, Coggeshall and Kumm[5] have shown that serum from monkeys harboring a chronic infection with *P. knowlesi* contains specific, protective antibodies since such serum hinders the progress of the disease in susceptible monkeys.

In this study Rhesus monkeys were immunized with formalin-killed *P. knowlesi*[6] parasites combined with a lanolin-like substance and paraffin oil containing killed and dried tubercle bacilli, for these substances proved to be effective in enhancing and sustaining antibody production with many antigens.[7] The antigen was prepared as follows: Citrated monkey blood containing large numbers of parasites were centrifugalized immediately after collection. The sediment was washed in 0.85 per cent. saline solution and suspended in 0.85 per cent. saline solution containing 0.1 per cent. formaldehyde. The suspension was kept in the refrigerator over night and washed three times with 0.85 per cent. saline solution. The sediment after the third washing was used for the unconcentrated vaccine. Concentrated vaccine was prepared by suspending the sediment of the third washing of formalinized red blood cells in 0.85 per cent. saline solution. After 48 hours in the cold the suspension was centrifugalized lightly and the reddish-brown cell-stroma-parasite layer was collected. The centrifugalization and the collection of parasitized cells was repeated several times. The final suspension contained large numbers of parasites and ghosts of red cells and very few normal red blood cells, whereas the residual red blood cell layer contained few parasites.

A water-in-oil suspension of the antigen in paraffin oil containing killed and dried tubercle bacilli was made with the aid of "Falba,"[8] the proportion of antigen, oil and "Falba" being 2:2:1 (Table 1).

[1] Manuscript completed September, 1944. See preceding paper.

[2] This study was aided by a grant from the John and Mary R. Markle Foundation.

[3] M. D. Eaton and L. T. Coggeshall, *Jour. Exp. Med.*, 70: 141–146, 1939.

[4] L. T. Coggeshall, *Medicine*, 22: 87–102, 1943.

[5] L. T. Coggeshall and H. W. Kumm, *Jour. Exp. Med.*, 68: 17–27, 1938.

[6] The strain of *P. knowlesi* was received through the courtesy of Dr. Johannes H. Bauer, of the International Health Division of the Rockefeller Foundation, to whom we are indebted for helpful advice and suggestions.

[7] (a) J. Freund and K. McDermott, *Proc. Soc. Exp. Biol. and Med.*, 49: 548–553, 1942; (b) K. Landsteiner and M. W. Chase, *Proc. Soc. Exp. Biol. and Med.*, 49: 688–690, 1942; (c) M. W. Chase, *Proc. Soc. Exp. Biol. and Med.*, 52: 238–240, 1943; (d) L. M. Kopeloff, S. E. Barrera and N. Kopeloff, *Am. Jour. Psychol.*, 98: 881–902, 1942, L. M. Kopeloff and N. Kopeloff, *Jour. Immun.*, 48: 297–304, 1944; (e) E. A. Kabat and M. H. Boldt, *Jour. Immun.*, 48: 181–183, 1944; (f) J. Freund and M. V. Bonanto, *Jour. Immun.*, 48: 325–334, 1944; (g) W. F. Friedewald, Science, 99: 453–454, 1944; (h) J. Freund and A. W. Walter, *Proc. Soc. Exp. Biol. and Med.*, 56: 47–50, 1944.

[8] Falba is said to be a mixture of oxycholesterins and cholesterins derived from lanolin (manufactured by Pfaltz and Bauer, Inc., New York, N. Y.).

TABLE 1
COMPOSITION OF VACCINE AND SCHEDULE OF INJECTIONS

Experiment	Monkey	Dose	Materials injected per monkey: Volume cc	Parasites billion	Red-cells billion	"Falba" cc	Paraffin oil cc	Killed dried tubercle bacilli mgm	Sites injected	Days between: Vaccinations	1st dose and challenge
1	15*	1st	2.26	4.5	15.4	.45	0.9	.45	3		
		2nd	7.5	3.0	12.6	1.5	3	1.5	3	46	
		3rd	7.5	14.7	25.9	1.5	3	3.0	3	28	118
	16*	1st	12.5	5	20.8	2.5	5	2.5	5		
	17*	2nd	7.5	14.7	25.9	1.5	3	3.0	3	28	71
	18*	1st	7.5	14.7	25.9	1.5	3	3.0	5		
		2nd	15.0	15	45.6	3	6	2.5	5	56	92
2	19†	1st	2.5	16‡	?§	0.5	1	2.5	5		
	20†	2nd	5.5	9.9‡	?§	1.1	2.2	1.4	5	52	86
	21†										

* Unconcentrated vaccine.
† Concentrated vaccine.
‡ Estimated from the red cell and parasite counts made on the unconcentrated blood.
§ Unknown.

Details of the procedure for making water-in-oil emulsions may be found in a previous paper.[7]

Each dose of vaccine was divided into three or five equal portions and injected into the subcutaneous tissue of the axillae, back of neck and groins. Subsequently monkeys so vaccinated (Table 1) and unvaccinated monkeys were infected by the intravenous injection of 2 to 3 cc of heparinized blood from an infected monkey which had been made a carrier by treatment with quinine.

Two experiments were carried out. In Experiment 1, there were 2 controls and 3 immunized monkeys; in Experiment 2, 6 controls and 4 immunized monkeys.

The parasite counts are shown in Tables 2 and 3. Table 2 contains the data on the unvaccinated monkeys of Experiments 1 and 2. It also includes 6 monkeys which received intravenous injections of 2 to 3 cc of blood from the carrier monkey, though

TABLE 2
PARASITEMIA IN NON-VACCINATED MONKEYS
PARASITES PER 100 RED BLOOD CELLS

Days after infection	Monkey number: 1	2	3	4	5	6	7	8	9	10	11	12	13	14
1	0	–	–	–	0	–	0	0	–	–	–	–	–	–
2	0	–	–	–	–	–	0	0	0	0	0	0	0	0
3	0	–	0	.4	0	–	0	0	–	–	–	–	–	–
4	0	–	.3	.3	0	–	0	0	0	0	0	0	0	0
5	0	0	–	–	–	0	.1	÷	0	0	0	0	0	0
6	0	0	14	10	.8	0	÷	.3	0	0	0	.1	0	0
7	0	÷	57K	53K	2	0	2	6	÷	0	0	÷	÷	.1
8	0	÷			9	0	40K	37K	1	.5	.1	.1	2	.5
9	0	1			D	0			10	.7	.7	8	8	3
10	÷	7				0			4	3	6	25K	24K	6
11	2.4	10				0			9K	9K	D			8
12	10.2	24K				0								5
13	36.0D					1.5								6
14						D								4
15														2
16														.3
17														0
18														.9
19														1
20														.4
21														0
22														0
23														0
24														–
25														0
26														0

– Not done.
÷ One parasite in more than 1000 r.b.c.
K = Killed.
D = Dead.
Experiment 1: Monkey Nos. 7, 8.
Experiment 2: Monkey Nos. 9, 10, 11, 12, 13, 14.
Monkey Nos. 1 through 6 were infected with the blood of the carrier but not simultaneously with animals in Experiments 1 and 2.
Autopsy: No tuberculosis: Monkey Nos. 1, 2, 3, 4, 7, 9, 11, 12.
Tuberculosis: Monkey Nos. 5, 6, 8, 10, 13.

they were not infected on the same days as the monkeys in Experiments 1 and 2. These animals may also be considered controls since Coggeshall and Eaton[8] demonstrated that the number of parasites introduced influences only the length of the prepatent period. Once the parasites become demonstrable, the progress of the disease is almost uniform, with death occurring usually on the 3rd to 6th day of the parasitemia. Table 2 shows that in the unimmunized animals parasites were first found in thin blood smears from 3 to 8 days after infection, and thereafter multiplied rapidly. Four of the 14 monkeys died with acute malaria. Nine monkeys were killed (to recover parasites for vaccine production). One unimmunized monkey survived after an infection which at its peak showed 8 parasites per 100 r.b.c. Mulligan and Sinton observed that 1 of 120, and Coggeshall and Kumm[5] 1 of 70 monkeys recovered spontaneously from an infection with *P. knowlesi*.

TABLE 3

PARASITEMIA IN VACCINATED MONKEYS
PARASITES PER 100 RED BLOOD CELLS

Days after infection	Unconcentrated vaccine			Monkey number	Concentrated vaccine		
	15	16	17	18	19	20	21
1	0	0	0	–	–	–	–
2	0	0	0	0	0	0	0
3	0	0	0	–	–	–	–
4	0	0	0	0	0	0	0
5	0	0	.1	0	0	0	0
6	÷	0	.1	0	0	0	0
7	1	0	.2	0	0	0	0
8	10	0	3D	0	0	0	0
9	4	0		0	0	0	0
10	8	.1		0	0	0	0
11	5	0		.1	0	.1	.1
12	6	.6		.4	0	.4	0
13	2	2		.5	0	1	.2
14	.7	2		.5	0	.6	1
15	.3	1		2	0	1	.5
16	.1	.4		2	0	.6	.6
17	.2	0		.5	0	.3	.1
18	.1	.1		2	0	.4	0
19	.2	0		6	0	.7	0
20	0	0		3	0	.3	0
21	.1	0		.2	0	.2	0
22	0	0		0	0	0	.1
23	.2	0		0	0	0	0
24	÷	.1		–	–	–	–
25	0	0		0	0	0	0
26	.1	0		0	0	0	0

Experiment 1: Monkey Nos. 15, 16, 17.
Experiment 2: Monkey Nos. 18, 19, 20, 21.
Monkey No. 15 died with extensive tuberculosis on the 69th day after infection. From the 27th to the 68th day it was examined for parasitemia on 21 different days. Parasites were found on 9 days, being 1 in 1,000 or more r.b.c. On the 68th day, one parasite was found in more than 1,000 r.b.c.
Monkey No. 16 died of tuberculosis on the 49th day after infection. From the 27th to the 49th day it was examined for parasitemia on 15 different days. No parasites were found.
Monkey No. 17 died in acute respiratory distress while a blood smear was being taken. Nine tenths of the cut surface of the lungs were involved in tuberculosis.

Table 3 shows the parasite counts in immunized animals. In one of 7 monkeys parasites were not found in thin blood smears. In the other six animals there were only 1, 1, 2, 3, 6 and 10 parasites per 100 r.b.c. at the peak of infection. Subsequently the parasites decreased until none were demonstrable in thin smears.

Although the number of monkeys in the experiments was small the difference between the course of infection in vaccinated and non-vaccinated monkeys seems significant considering the high virulence of *P. knowlesi* for the Rhesus monkey.

Palpable masses were found in the subcutaneous tissue at the sites of injection in all vaccinated monkeys. These masses did not ulcerate through the skin in the three monkeys which showed no reaction to tuberculin (P.P.D.). However, it may be noted that one of these monkeys (No. 15) showed extensive tuberculosis at autopsy 85 days after its negative reaction to P.P.D. Abscess formation and ulceration through the skin were observed in the four monkeys which reacted to P.P.D. Two of these animals died and showed extensive tuberculosis at autopsy.

CONCLUSION

The injection of formalin killed *P. knowlesi* parasites combined with a lanolin-like substance and paraffin oil containing killed tubercle bacilli modifies parasitemia and prevents fatal infection with *P. knowlesi* in Rhesus monkeys.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

METHOD FOR THE DETECTION OF INDOLE[1]

THE detection of indole is a matter of importance in a number of different fields. For instance, principal uses of this test are (1) the differentiation of bacteria, some of which characteristically produce indole while others do not[2]; (2) the detection of spoilage of foods[3]; (3) the demonstration of the

[1] Contribution from Department of Health, City of New York.

[2] Am. Pub. Health Assoc. Standard Methods of Water Analysis. 8th ed. 1936.

[3] Clarke, Cannon, Coulter, Goodman, Greene, Milsted, Vandaveer and Wildman, *Jour. Assoc. Official Agr. Chem.*, 20: 475, 1937; *Chernoff, Ind. Eng. Chem.*, Anal. Ed., 12: 273, 1940.

presence of indole producing organisms in milk,[4] food and water as evidence of pollution; and (4) detection and determination of indole in blood,[5] urine,[6] and feces[7] for clinical diagnosis.

The most widely used test for the detection and determination of indole is its reaction with *p*-dimethylaminobenzaldehyde. This reaction, introduced by Ehrlich[8] and modified by Kovács,[9] has achieved official standing by being adopted by the American Public Health Association as one of the tests for the differentiation of *E. coli* from *A. aerogenes*. This direct reaction, however, is not specific for indole, for an entire series of compounds, such as aldehydes, also give colored compounds, nor is it sufficiently sensitive.[10] Because of this lack of sensitivity it is necessary to isolate the indole before applying the test reaction. This has been done by extraction with immiscible solvents, by steam distillation and by dialysis.

p-Dimethylaminobenzaldehyde-trichloroacetic Acid Test

The following method, which can detect directly 1 microgram of indole in 10 ml of test solution or 1 part of indole in 10 million of solution or by an extraction modification can detect 1 microgram in 100 ml of test solution or 1 part per 100,000,000, is relatively simple, rapid and sensitive.

Reagents: (A) Dissolve 10 grams of trichloroacetic acid in 30 ml of chloroform. Add 2 grams of *p*-dimethylaminobenzaldehyde. Transfer to a separatory funnel and saturate by shaking with concentrated hydrochloric acid. This reagent is stable for at least 2 months. (B) Dissolve 10 grams of trichloroacetic acid in 30 ml of chloroform. Add 2 grams of *p*-dimethylaminobenzaldehyde. Add 0.5 ml of acetic anhydride and 2 drops of concentrated sulfuric acid. This reagent is stable for at least 2 days. It is more sensitive than the reagent (A) above.

Detection: Transfer 10 ml of the culture or 10 ml of the sample solution to a separatory funnel. Add 2 ml of chloroform. Shake vigorously and allow to stand for 5 minutes. Swirl to make sure the chloroform layer collects at the bottom. Filter through a filter wetted with chloroform into a small test-tube or insert a pledget of cotton into the stem of the separatory funnel and draw off the chloroform layer directly into a test-tube. Add 2 drops of either reagent listed above and hold in warm water (50° C.) for 0.5 minute. A pink, to rose or red, color is positive for indole. A straw color or light yellow is negative for indole.

Instead of the chloroform extractant, 2 ml of a mixture of equal volumes of chloroform and carbon tetrachloride may be used.

Determination: Shake out twice with chloroform or with the mixed solvent. Filter into a colorimeter tube or a 10 ml volumetric flask as directed above. Make up to volume with the mixed solvent or chloroform and compare against standards treated the same way.

Discussion: Only for very low concentrations of indole is the chloroform extraction necessary. In most instances, especially in cultures after 24 hours incubation, the reagents may be added directly to the culture to be tested. The color forms at the interface or is extracted by mild shaking and is found in the bottom chloroform layer.

The addition of 5 ml of acetic anhydride to reagent (A) does not seem to alter its properties but if acetic acid is used instead of hydrochloric acid, it does not give the reaction. Dilution with half its volume of carbon tetrachloride reduces its sensitivity. Reagent (B) without acetic anhydride will work only if freshly prepared. It deteriorates rapidly.

With low concentrations of indole of the order of 1 to 0.1 micrograms per ml of chloroform, the pink color fades out rather rapidly with reagent (A). With higher concentrations, the color does not fade out even on standing 24 hours. The color formed with reagent (B) does not fade even after standing for hours. When carbon tetrachloride is used as part of the solvent mixture, the color comes up fast but fades out rapidly. It lasts longest with reagent (B) but is weak in intensity.

The reagent is affected by water, consequently when it is shaken with a culture or with aqueous solutions it loses in sensitivity. For greatest sensitivity it is necessary to use the extraction method as detailed above.

Experimental: The solutions of indole were prepared by dissolving 1.00 gram of indole in 95 per cent. alcohol, transferring to a 100 ml flask and diluting to volume with the same solvent. Suitable aliquots were withdrawn and diluted with water in volumetric flasks so that concentrations of the order of 0.01 to 1 microgram per ml of test solution were obtained. These solutions were used to establish the order of sensitivity mentioned previously.

The bacteriological method was tested on 167 cultures of which 46 were *E. coli* cultures (10 specimens), 8 were salmonella cultures (2 specimens), 7 were Sonné cultures (1 specimen), 4 were aerogenes cul-

[4] Damm and Bartram, Molkerei-Ztg. (Hildesheim) 50: 714, 1936; *Chem. Abstracts*, 30: 6836, 1936.
[5] Mazzocco, *Rev. soc. argentina biol.*, 11: 31, 1935; *Compt. rend. soc. biol.*, 119: 699, 1935.
[6] Vaughan, *Proc. Soc. Exp. Biol. and Med.*, 29: 623, 1932.
[7] Bergeim, *Jour. Biol. Chem.*, 32: 17, 1917.
[8] Ehrlich, *Deut. med. Wochschr.*, 1901, 1.
[9] Kovács, *Z. Immunitäts.*, 55: 311, 1928.
[10] Lehr, *Centr. Bakt. Parasitenk.* Orig. I Abt. 108: 209, 1928.

tures (1 specimen), 84 were cultures of sewage (6 specimens), and 22 were uninoculated blanks.

Regulation methods were used for the inoculation of a tryptone culture medium. Cultures were withdrawn from incubation in the first experiments at the end of 1, 2, 3, 4, 5, 6 and 24 hours and were tested for indole. Later experiments on cultures were performed after incubation at 37° C. after 2, 3, 4 and 24 hours.

Results: The inoculated specimens of *E. coli* yielded positive results in 7 out of 8 cases at the end of 3 and 4 hours incubation and in the 8th at the end of 24 hours incubation. However, 2 specimens were negative throughout. Four out of 8 were positive for indole at the end of 2 hours incubation. Mastafa[11] noted that a medium made by peptic digestion of entire hogs' stomachs could be used for the detection of indole at the end of 2 hours incubation. The Sonné, salmonella and aerogenes specimens were negative for indole production as expected. Except for trace results, the experiments with raw sewage, and treated sewage, obtained from a large city sewage treatment works, diluted 1:10, 1:100, 1:1000, 1:10,000, and 1:100,000 and undiluted, were negative or at best yielded a trace result for indole, at the end of 5 hours incubation, with the exception of two undiluted raw sewage specimens which yielded positives at the end of 5 hours incubation. However, every dilution of raw sewage yielded a strong positive at the end of 24 hours. Partially treated sewage yielded positives in dilutions up to 1:100 and settled sewage only when undiluted.

Acknowledgment: The authors wish to thank Miss Nancy Ferranti for her assistance with the bacteriological work.

SUMMARY

A simple and sensitive method for the detection of indole in cultures, urine, sewage, etc., is presented. The indole is extracted with chloroform and is then treated with a modified Ehrlich reagent. The possibility of use for the detection of water pollution is indicated.

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AN INSTRUMENT FOR THE RAPID MIXING OF FLUIDS IN SMALL TUBES[1,2]

IN the preparation of serial dilutions for purposes such as the titration of viruses and immune sera, the material in question is transferred to a tube containing diluent. Mixing is usually then effected by drawing the fluid to and fro in a pipette several times before transfer is made to another tube for the next dilution. When many dilutions are to be made, this process of mixing is not only time-consuming but physically wearing. In the contemplation of a large series of studies[3,4] on the titration of serum antibodies inhibiting the hemagglutinative action of the influenza virus,[5] a simple means was found for obviating much of the tedium and loss of time. This consists in the use of an electric massage vibrator[6] of the type shown in Fig. 1, which is a drawing taken from

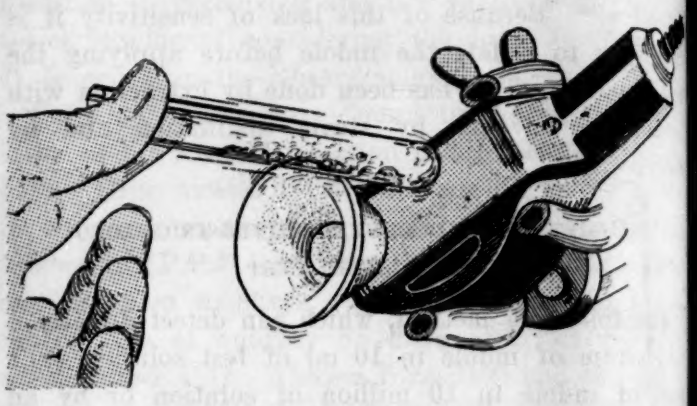

FIG. 1. A drawing of the vibrator and the mixing of fluid in the tube based on the tracing of a photograph.

a photograph. The motor mechanism is mounted inside a closed metal chassis from which there projects a short rod. The rod is the vibrating part, and for it there are provided several rubber attachments. The best of these for the purpose is the bell-shaped one illustrated in Fig. 1. The instrument is fixed to the table with an iron clamp, set going and the tube containing the materials to be mixed is held against the edge of the bell. As shown in Fig. 1, the mixing action is highly effective. Some trial is necessary, however, for finding the best point and the proper angle for application of the tube.

Repeated tests have shown that mixing as good as that with a pipette can be obtained by holding the tube against the machine with the left hand during the period in which the right hand is selecting the

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[2] This work was aided by the Commission on Influenza and the Commission on Epidemiological Survey, Board for the Investigation and Control of Influenza and Other Epidemic Diseases in the Army, Preventive Medicine Service, Office of the Surgeon General, United States Army. The work was aided also in part by a grant to Duke University from Lederle Laboratories, Inc., Pearl River, N. Y.

[3] I. W. McLean, Jr., D. Beard, A. R. Taylor, D. G. Sharp and J. W. Beard, *Jour. Immunol.*, in press.

[4] *Idem.* In preparation.

[5] G. K. Hirst, *Jour. Exp. Med.*, 75: 49–64, 1942.

[6] Allover A.C. Vibrator, Allover Mfg. Co., Racine, Wis.

[11] Mastafa, *Compt. rend. soc. biol.*, 124: 450, 1937.

new pipette for the next transfer. In a routine experiment a single operator prepared 823 individual dilutions in a total elapsed time of 114 minutes, an average of 7.2 per minute.

JOSEPH W. BEARD

A NEW COLORIMETRIC REAGENT FOR TITANIUM

IN a report on their investigation of disodium-1,2-dihydroxybenzene-3,5-disulfonate [$C_6H_2(OH)_2(SO_3Na)_2 \cdot H_2O$] as a reagent for the colorimetric determination of iron, Yoe and Jones[1] observed that it gives an intense yellow solution with Ti^{+4}. Their preliminary observation indicated the sensitivity to be about 1 part of titanium in 200 million parts of solution when comparisons are made in 50 ml, tall-form Nessler cylinders. This observation has been substantiated.

The color intensity of the titanium complex is independent of acidity over the range pH 4.3–10, the color does not change in intensity or tint over periods of several months, and it obeys Beer's law over the useful range of concentration.

The number of interfering ions is small. Aluminum, calcium and tungsten reduce the intensity of the color; this can be largely overcome by adding an excess of reagent. Iron, vanadium and uranium develop colors with the reagent, but only the first is commonly encountered. The purest available reagents used for opening up samples contain sufficient iron to give an off-tint color to the titanium complex. The iron interference may be eliminated by buffering the solution at pH 4.7 with acetic acid and sodium acetate in a 1:1 molar ratio and adding 50 mg of sodium hydrosulfite per 100 ml of solution. Under these conditions the iron is reduced to the ferrous state and gives no color with the reagent, and hydrosulfite solutions show no turbidity for 20 minutes.

If titanium and iron are both to be determined, this may be done with the same solution. Add the reagent (about 0.1 g), adjust to pH 4.7, measure the absorbency (-log of transmittency) at 560 mμ (the maximum for the iron complex at pH 4.7), then reduce with sodium hydrosulfite and measure the absorbency at 410 mμ. The amount of iron and titanium may be determined from previously prepared graphs.

A more extensive report on the use of this reagent for the colorimetric determination of titanium will be published elsewhere in the near future.

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DISCUSSION

THE PHYSICAL CHEMISTRY OF COLOR VISION

THE manifold and long-continued research work of Selig Hecht and his collaborators on the nature of vision and visual response to light has culminated or converged, in one aspect, that of color vision, in a rather radical transformation of the trichromatic theory initiated by Thomas Young, developed by Clerk-Maxwell and Helmholtz, disputed by Hering and exploited practically in various processes of color photography. This transformation is proposed by Dr. Hecht most concisely, for present reference, in his paper on "The Development of Thomas Young's Theory of Color Vision" and very important confirmatory and extensory investigations are given in a more recent paper by J. Mandelbaum and E. U. Mintz.[1]

The basic nature of the transformation is evident on comparing the accepted trichromatic "excitation" (or sensitivity) curves of Koenig, Abney, Wright and others[2] and those introduced by Hecht. These, instead of differing widely throughout the spectrum, are close together, and have maxima in the yellow-green part of the spectrum near the region of maximal photopic visibility; the shape of the curves varies but little one from another, and from the photopic-visibility curve.

As pointed out by Mandelbaum and Mintz, "The virtue of Hecht's formulation is that for the first time much of the data relevant to color vision is included."

Hecht has stated the difficulties of the physico-technical theory[3] for the physiological (and chemico-physical) picture as follows: referring to the accepted values of the excitation curves adopted by the American Optical Society and the visibility curve (of Gibson and Tyndall) he says: "These are all unimpeachable facts. But they make it extremely difficult to formulate a physiological picture of what they mean, especially in the matter of treating the excitation curves or the *Grundempfindungen* as the real physiological primaries which must accomplish what Thomas Young's notion expects them to." In face of the difficulties, Hecht has proposed, as he himself, I think too modestly, has said, a set of "Variations on a Theme by Thomas Young." It is, incidentally, a part of the intention of this note to suggest that there has been so radical a transformation of the theme as to

[1] *Ind. Eng. Chem., Anal. Ed.*, 16: 111, 1944.
[1] *Am. Jour. Ophthal.*, 24: 1241, 1941.
[2] *Jour. Ophthal. Soc. America*, 20: 231, 1931.
[3] Thus to designate the current theory.

create a new symphony. This because, if the relation of Hecht's suggestions to those that follow be in any way grounded, the dominant triune or Pythagorean threefold character of the original theme is in course of replacement by a numerical discontinuum of order $\leqq$ and generally < 5, but probably not essentially and always 3. The meaning of this will be clear, when it is allowed that for protanopes the number of color-processes could be $=1$ or >1, but not $=2$,[4] for deuteratopes 2 or 3, and for normal polytopes, 3 or greater.[5] This is one consequence of the concept of closely overlapping curves—conforming to Hecht's conclusions, but admitting statistical variates. Mandelbaum and Mintz suggest:

> From a phylogenetic and from a photochemical point of view, the concept of closely overlapping curves is more probable than is that of the widely separated curves of classical formulation. Their differences in sensitivity may well be due to slight differences in their absorption spectra, for in the case of both visual purple and visual violet the spectral sensitivity is determined by the absorption spectrum. Such slight differences in spectral absorption could easily be accounted for by small molecular rearrangements, or the simple addition of a methyl or ethyl group.

What we wish to point out, as a mere suggestion for possible further prosecution by physiologists and biochemists, is that well-ordered and physico-chemically controlled modifications of the absorption spectrum exist in dyes of many types, which would permit the closely overlapping curves of the Hecht theory to arise for the same dye as well as by essential, though small, chemical modifications of a dye molecule. Such alterations can be determined by molecular aggregations of a reversible character, controlled by the parameters of the thermodynamic environment. We propose to group such variations under the general term, allelochromy,[6] and may mention two principal directions in which conditions for fulfilment of the closely overlapping trinity—or multeity—of sensitivity curves might be sought. These are (1) spectral absorption variation by dimerization[7] and (2) by heteropolymerization of dyes in lyotrope mesophase formation.[8] These are quite distinct processes, though liable to appear with the same dye molecules, and for the good reason that both depend upon the possibility of sterically unimpeded side-by-side parallel coherence, in the first case in pairs of two ions, in the second, in a species of one dimensional or filamentous quasi-crystallization of large numbers of like dye molecules acting as a unit.

Spectral variations in the first case can be produced by variation of concentration and medium. A pronouncedly amphipathic substance,[9] such as the nucleoprotein of nerve-fiber, should be ideal in respect of such control. *Per contra,* the other state of ordering also deserves consideration. While the striking spectral absorption of the mesophase of 1,1′-diethyl-2,2′-cyanine chloride, discovered by E. E. Jelley[10] and, independently, by G. Scheibe,[11] has concentrated attention on this narrow band type from symmetrical molecules, it deserves to be noted that with asymmetrical dye molecules of the cyanine dye family, cooperative spectra giving absorption curves of much broader form may be obtained, and, again, by small variations of the proportions of reversibly heteropolymerized ions, allow controlled variation of the absorption.[12] In this case, with *two* not too dissimilar but isomorphous dye ions, A and B, *three* packets of A, AB and B could give curves of near overlap. The writer has commented elsewhere[12] on the analogies of the combinations such dye ions can make with proteins, with the spatial arrangements made probable for thymo-nucleic acid molecules.[13] The conditions by which, in these multi-molecular assemblages, excitation-conductance becomes possible, also would seem to deserve attention in connection with the theory of vision.

It is an interesting thought that while the widely separated curves of the classical trichromatic theory have well served the mimetic development of color photography, operation on the basis of such closely overlapping curves as those proposed by Hecht would probably have been technically impossible, or, in any case, impractical.

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PROPOSED UNIT FOR HIGH VACUUM

F. H. TOWNSEND[1] suggests a new unit for the measurement of vacuum which has the virtue of expressing increasing vacuum—decreasing pressure—in terms of

[4] Fractional values referring to statistical variations of the distributions of operative processes (1, 2, 3, 4 or 5) in rod and cone populations.

[5] Just as in multicolor printing, it could be a matter of economy *vs.* efficacy whether two, three or more components were employed. But, for vision, it is rather the (statistical) frequency of certain dye molecules in the populations of rods and cones.

[6] *Cf.* allelomorphy and signifying alternative or contrasting coloration.

[7] S. E. Sheppard and A. L. Geddes, *Am. Chem. Soc.*, 66: 1995, 1944, Part IV.

[8] S. E. Sheppard, *Reviews of Modern Physics*, 14: 303–340, 1942.

[9] S. E. Sheppard and A. L. Geddes, Amphipathic Character of Proteins and Certain Lyophile Colloids as Indicated by Absorption Spectra of Dyes, *Jour. Chem. Phys.*, 13, 63, 1945.

[10] E. E. Jelley, *Nature*, 138: 1009, 1936.

[11] G. Scheibe *et al.*, *Naturwiss.*, 25: 75, 1937.

[12] S. E. Sheppard, SCIENCE, 93: 42, 1941.

[13] K. Linderstrom-Lang, *Trans. Farad. Soc.*, 31: 324, 1935.

[1] *Nature*, 155: 545, 1945.

increasing positive numbers. At present we write 10^{-1}, 10^{-2}, 10^{-3}, . . . mm of mercury; or we use 10^{-3} mm, the micron, as the basic unit for measuring vacuum, and write 1, 0.1, 0.01, . . . , micron.

In Townsend's proposed system, the reciprocal of the negative power of 10, expressing pressure in millimeters of mercury, is multiplied by 10, thus resulting in positive, integral numbers of two figures. In this system, 10^{-1}, 10^{-2}, 10^{-3}, . . . , become 10, 20, 30, . . . vacuum units. This is similar to the method of expressing relative sound intensity levels, the reference level for vacuum being 1 mm. There is logic in using approximately 1 mm as the division point between pressure and vacuum. In its practical application, the range of values ordinarily used would lie between 10 and 70 vacuum units.

The suggestion appears practicable and logical. This note is written to bring it to the attention of American scientists and technologists who may not have access to *Nature*.

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THE OXIDATION OF BILIRUBIN BY PEROXIDASE

PEROXIDASE, in the presence of hydrogen peroxide, is known to oxidize phenols and aromatic amines and iodides. Milk peroxidase oxidizes nitrites and tryptophane.[1] Recently we have discovered that peroxidase oxidizes bilirubin to biliverdin. This reaction takes place in a narrow zone around pH 7.4.

It has been generally assumed that *in vivo* hemoglobin is broken down to biliverdin and that the biliverdin is then reduced more or less completely to bilirubin. However this may be, we wish to point out the possibility that bilirubin may be converted to biliverdin in the liver by action of peroxidase and peroxide. In 1934 Schreus and Carrie[2] observed that when liver brew was incubated with hemoglobin between pH 7 and 8, a blue-green pigment was formed. This was probably biliverdin. The formation of this substance was shown to be inhibited by catalase and favored when the digests were kept at 70°, at which temperature liver catalase was destroyed. We assume that the inhibitory effect of catalase was due to its destruction of hydrogen peroxide.

We wish to acknowledge our thanks to the Rockefeller Foundation for financial assistance.

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[1] S. Thurlow, *Biochem. Jour.*, 19: 175, 1925.
[2] H. T. Schreus and C. Carrie, *Klin. Wochenschr.*, 13(2), 1670, 1934; *Med. Welt*, 9: 1135, 1935; quoted from *Chem. Abstracts*, 31, 6310, 1937.

THE IMPORTANCE OF DEGENERATIVE CHANGES IN LIVING ORGANISMS

VARIATIONS that reduce the amount and rate of growth in long-inbred lines of maize appear in the form of dwarf plants, narrow leaves, crooked stalks, reduced chlorophyll and plants that seem normal but are delayed in flowering and maturing. All the mutations so far observed are either neutral or disadvantageous to the organism. Unfavorable types such as these would be expected to be eliminated by natural competition, but when tested in hybrid combinations with the normal lines from which they originated, these degenerate individuals improve the performance of their offspring and definitely have advantage in survival. Increases over the normal, better parent range up to 104 per cent. in yield of grain, and up to 9 per cent. in height of stalk. This larger growth is made in less time.

Favorable mutations in plants and animals are extremely rare both under natural conditions and in the laboratory. The evidence indicates that they appear first in the heterozygous condition and segregate as unfavorable deviations from normal. Hereditary material is so highly developed and delicately balanced that changes of any kind usually result in a reduction of some kind. Ultimately these new alleles may be brought into equilibrium with the remaining gene complex in such a way as to promote better growth and survival. In this way evolution proceeds by first taking a step backward before going forward.

The fact that unfavorable characters appear so frequently and persist so long in many organisms indicates that they have survival value. These results, recently obtained and to be reported in detail, emphasize the need for caution before eliminating apparently degenerate individuals in plant and animal breeding practice as well as in any eugenic program.

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A REACTION OF ASCORBIC ACID WITH α-AMINO ACIDS

IN SCIENCE of May 25, 1945, Koppanyi, Vivino and Veitch described a red color reaction of ascorbic acid with various α-amino acids. Somewhat over a year ago we observed the development of a bright red color on and in surgical catgut (mainly collagen) when immersed in ascorbic acid solutions, and we believe the same reaction as described by those authors to be involved.

When raw or processed (heat sterilized) plain catgut strands were immersed in ascorbic acid solutions of 50 mg per cent. or more, and stored at room temperature, the strands were uniformly red in three

days. The "dyeing" of the gut coils, kept in upright stoppered test-tubes, appeared after one day on the top end, progressing toward the bottom. No reagent other than ascorbic acid was added, and heating did not accelerate the reaction. The red color was stable for several weeks, then turned gradually brown. The liquid stayed entirely clear and colorless.

This color development took place when the ascorbic acid was dissolved in absolute or in 95 per cent. U. S. P. ethanol, or in a mixture of ethanol-isopropanol 4:1, containing 5 per cent. water, not however in absolute methanol or in water. In the latter two solvents a brown discoloration of the gut was obtained, similar to, though weaker than that developing from the red color. With xylol as medium no visible reaction took place.

The alcoholic or alcoholic-aqueous systems employed cause gut to soften and swell (non-boilable catgut), the final stage being reached within one week. Collagen contains available amino groups whose number may be gradually increased when swelling. The fact that the color is found exclusively on the gut strands and not in the liquid media, suggests the reaction of the ascorbic acid to take place with insoluble reactive groups ($-NH_2$) of the protein, ruling out ammonia in the case of gut.

The oxidation of the ascorbic acid may be attributed to air oxygen, evidenced by the progression of the color from the top, near the liquid-air interphase, toward the bottom.

Koppanyi *et al.* have pointed out that the color is less stable in water than in alcohol (supposedly ethanol). This and the lesser stability of ascorbic acid in water and apparently methanol can explain the failure of the red color to appear in these solvents. In xylol gut does not swell nor soften (boilable catgut), and the solubility of ascorbic acid is low if not zero, hence no reaction takes place. Qualitative tests for ascorbic acid according to Szent-Györgyi (Merck Index 3955), carried out under same conditions for all systems, were positive after a three-day period in all solvents, except in methanol (destruction?) and in xylol (insolubility?). A number of other reducing agents or sugars did not cause this color development with catgut. The reaction of ascorbic acid with other proteins has not been studied, but may furnish some information as to their characteristics.

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HOMING, MIGRATION AND INSTINCT

PLATT and Dare, under the heading "The Homing Instinct in Pigeons" in your issue of April 27, 1945 (p. 439), express the belief that it is more reasonable to "explain the homing and migratory habits of birds by assuming that they use familiar landmarks, together with simple geographical, meteorological and ecological relationships rather than that they possess a new sense organ." They describe recent experiments in the homing of pigeons and point out that their results suggest that training and familiarity with landmarks are integral factors in the successful return of birds to their lofts.

While this opinion corroborates, in essence, the views expressed by earlier investigators of homing in pigeons,[1] it should be pointed out that the coupling of the words "homing" and "migration," as though they were different manifestations of a single phenomenon, appears to be unwarranted. Pigeons have been used often enough as "guinea-pigs" of the air in attempts to elucidate questions of migration (the physicist, Kelvin, being possibly the first to have done so) yet their very success as homing agents rests primarily on the fact that they are entirely devoid of any migratory instinct. One can not, in fact, argue legitimately from the homing habits of pigeons to the migratory ways of other species.

That the generalization made by Platt and Dare can not be upheld is evidenced in the first annual migrations of the young of numerous species of birds which undertake their initial fall journey without knowledge of landmarks or the chaperonage of adults. The young of our cowbirds, for instance, or European cuckoos, reach their predestined wintering grounds without either parental or foster-parental guidance, while certain species of the flightless penguins migrate annually by swimming from the antarctic to South America and back with infallible precision, through a murky ocean from which they are presumably incapable of getting bearings and on which there exist no landmarks.

On November 9, 1940, approximately a month after the last resident crow had gone south, I liberated 54 young crows of the year near Edmonton, Alberta, from the area on which they had been hatched and subsequently trapped as juveniles in July and August. They were merely held in a spacious flying cage during the intervening period; no adults were with them. By November 20 over 50 per cent. had been retaken, the furthest 250 miles southeast of the point of liberation on a line directly joining Edmonton and central Oklahoma, the wintering ground of 95 per cent. of Alberta crows. None of the birds recovered had deviated materially from this line and some of them were traveling at 50 miles per day, a remarkable rate for crows. The temperature was below zero F. and the ground blanketed with snow.

[1] *E.g.*, B. B. Riviere, Verhandlungen des VI. Internat. Ornithologen-Kongresses, Kopenhagen, 1926.

On this, their first southward migration, these birds were doing the opposite to homing: they were deserting their birthplace for a destination that could have been known to none of them and over territory on which not a single landmark could have raised familiar memories or been previously observed.

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SCIENTIFIC BOOKS

ELECTRICITY AND RADIO

Basic Electricity. By WILBUR L. BEAUCHAMP and JOHN C. MAYFIELD. viii + 312 pp. Illustrated. Chicago: Scott, Foresman and Company. 1943. $1.60.

Fundamentals of Electricity. By WILLIAM H. JOHNSON and LOUIS V. NEWKIRK. x + 212 pp. Illustrated. New York: The Macmillan Company. 1943. $2.00.

Fundamentals of Electricity. By CHARLES E. DULL and MICHAEL N. IDELSON. xx + 456 pp. Illustrated. New York: Henry Holt and Company. 1943. $2.00.

Fundamentals of Electricity. By MORTON MOTT-SMITH. 64 pp. Illustrated. Pittsburgh: Westinghouse Electric and Manufacturing Company and Science Service, Inc. 1943.

Electricity and Its Application to Civilian and Military Life. By CHARLES A. RINDE. xii + 466 pp. Illustrated. New York: Harcourt, Brace and Company. 1943. $2.50.

Prepare Yourself. By LAWRENCE F. TULEEN, GEORGE S. PORTER and ARTHUR HOUSTON. vi + 298 pp. Illustrated. Chicago: Scott, Foresman and Company. 1943. $.96.

Shop Job Sheets in Radio. By ROBERT NEIL AUBLE. Book I—*Fundamentals,* vi + 134 pp. Book II—*Service Problems,* x + 128 pp. Illustrated. New York: The Macmillan Company. 1944. $1.50 each.

Practical Radio and Electronics Course. By M. N. BEITMAN. (3 volumes) 368 pp. Illustrated. Chicago: Supreme Publications. 1943. $3.95.

AT the beginning of the present war, it was obvious that the armed forces would make wide application of the amazing developments of the past two decades in the field of electronics. It was also obvious very early in the conflict that there was a great lack of individuals trained to operate the new highly specialized and rather complicated equipment. This lack was partly caused by the failure of most educational institutions to afford proper emphasis to electronic devices and instruments in their laboratory courses. While courses in applied electronics had been introduced in many schools, the general attitude was that the work was not too important and consequently did not enjoy the wholehearted support of many of the educators.

Fortunately, once the new electronic equipment has been designed, engineered and built, its operation is relatively simple and can be entrusted to relatively unskilled personnel. The armed forces were then faced with the necessity of giving a training program to a large group from whom this operating personnel could be selected. For such training programs, most of the available texts were utterly unsuitable in that either the treatment followed the conventional lines of logical presentation common to most college-grade text-books or the treatment in the usual high-school text was too elementary. To fulfil the need, a number of texts were prepared all according to a definite specification as to the subject-matter, method of treatment, etc. The above listed texts were some of the books written for this purpose.

As would be expected from the fact that these books were designed to definite specifications, the treatment is similar in arrangement, scope, etc. In all these books, the subject of magnetism and electricity is treated before the subject of mechanics, with the result that the conventional treatment based on mechanics is not possible. Another result of this is that the scope of the numerical problems which can be assigned is definitely limited, particularly in not making it possible to stress the energy relations which exist in electrical circuits. Here again, since the books are designed to "train rather than educate," the loss is not too serious. In all the books, considerable emphasis is placed on the subject of electro-chemistry, as applied to primary and storage cells. The main difference between the various books lies in the selection of examples, photographs and diagrams, and in this respect all of the books are very good.

"Basic Electricity," by Beauchamp and Mayfield, and "Fundamentals of Electricity," by Johnson and Newkirk, are very similar in many respects and are definitely elementary in their treatment. The first of these books actually describes many experiments with detailed instructions on the making of the apparatus for the experiments. "Fundamentals of Electricity," by Dull and Idelson, is just slightly more advanced and more emphasis is placed on numerical problems and formulae. "Fundamentals of Electricity," by Morton Mott-Smith, was prepared by the Westinghouse engineers and looks rather more like a collection of reprints from popular magazine articles than a text-book. It is, however, especially good in containing a large number of excellent sketches and photographs. "Electricity and Its Ap-

plication to Civilian and Military Life," by Rinde, while written to the same specifications, is perhaps the most advanced text of this group, and it also contains many excellent diagrams and photographs. As indicated by its title, the main emphasis in this book is on electricity and its treatment of mechanics is very brief indeed although probably adequate for the purpose.

"Prepare Yourself," by Tuleen, Porter and Houston, is a little different from the others in that it is an elaborate laboratory manual. The various standard physics experiments, beginning with mechanics, heat, sound, light, magnetism and electricity are treated in a rather unconventional manner. The student is guided through a series of simple experiments, many with common household devices, and, by means of questions and suggestions in the text, he is asked to ascertain the physical facts. Space in the book is provided for filling in the answers. Consequently, when the student has gone through this book and performed most of the experiments, he might not have a knowledge of formal physics, but he will know as many facts and will probably have a better idea of what is really aimed at in laboratory work than many students who have gone through the more formal training. The level of this book is about that of the last year of high school or possibly junior college grade. Emphasis on the idea that experiments are made to find the facts is stressed throughout the book. This factor is all too frequently neglected, particularly in some high-school physics laboratories.

The last two books, which are in a different class from those mentioned, are: "Shop Job Sheets in Radio," books 1 and 2, by Auble, and "Practical Radio and Electronics Course," Volumes 1, 2 and 3, by Beitman. The first of these is again a laboratory manual of experiments in elementary electricity with emphasis on radio and, in the second, specific experiments on radio sets. The second book, "Practical Radio and Electronics Course," is a little more elaborate and is more of a text. The three volumes of this cover fundamentals of "Radio and Electronics"; "Receivers, Transmitters and Test Equipment," and "Applied Electronics and Radio Servicing." A large number of photographs of commercial equipment and service men's kinks are found in these books. The treatment, however, is primarily such as would be useful for a trade school in that no effort is made to present logical treatment considered essential to the student of any science. There are, however, many useful kinks described in these books so that even a professional physicist would no doubt find many ideas of use to him.

It is possible that after the war the books above described will go out of print and be no longer available. Nevertheless, there is a definite place for books of this character which should be useful for the individual who wishes to go on in electricity a little beyond the conventional high-school treatment but who would not care for some reason or other to study the more conventional treatment covered in the college-grade text-books. Authors of college-grade text-books would do well to look over these various books and use some of the ideas and illustrations to liven up their own texts, which often become rather dull reading because of the author's insistence on rigorous logical treatment. All too frequently when so-called practical ideas or applications are discussed, the treatment is in the nature of an apology.

While none of these books by itself would be suitable for a formal course in physics either in the elementary schools or advanced schools, they would all be very suitable to use as supplementary texts. The treatment, which is slightly more advanced than usual high-school treatment, would make it possible for the more intelligent high-school student to advance a little beyond conventional high-school treatment. Also, the books should assist the students of college grade in showing more practical illustrations than are usually given in a conventional college-grade text. In every case, the various authors are to be congratulated on having produced these books under what must have been considerable pressure to complete them in the shortest possible time. The style of writing in all of them is easy to read and follow. With the exception of the book by Morton Mott-Smith, the pages and type are well arranged for easy reading.

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BOOKS RECEIVED

BAILEY, ALTON E. *Industrial Oil and Fat Products.* Illustrated. Pp. x + 735. Interscience Publishers. $10.00. 1945.

GEORGIEV, ALEXANDER M. *The Electrolytic Capacitor.* Illustrated. Pp. xii + 191. Murray Hill Books, Inc. $3.00. 1945.

HERZBERG, GERHARD. *Infrared and Raman Spectra.* Illustrated. Pp. xiii + 632. D. Van Nostrand Company, Inc. $4.50. 1945.

KEENAN, JOSEPH H. and JOSEPH KAYE. *Thermodynamic Properties of Air.* Pp. iii + 73. John Wiley & Sons, Inc. $2.25. 1945.

SIMONDS, HERBERT R., M. H. BIGELOW and JOSEPH V. SHERMAN. *The New Plastics.* Illustrated. Pp. xii + 320. D. Van Nostrand Company, Inc. $4.50. 1945.

SMITH, JAMES G. and ACHESON J. DUNCAN. *Sampling Statistics and Applications.* Illustrated. Pp. xii + 498. McGraw-Hill Book Company. $4.00. 1945.

WILSON, CHARLES M. *New Crops for the New World.* Illustrated. Pp. viii + 294. The Macmillan Company. $3.50. 1945.

YOUNG, C. B. F. *Chemistry for Electroplaters.* Illustrated. Pp. vi + 205. Chemical Publishing Co., Inc. $4.00. 1945.